

A Scandal Ameliorated

THE new British Government has responded sensibly and quickly to the recommendation of the Dainton committee on libraries that there should be in future a unified library service for the United Kingdom. The intention, made public last week in a White Paper (*The British Library*, HMSO, Cmnd 4572. 1s 6d), is that the British Museum Library, the National Central Library (which aims at providing a centralized service for university and public libraries), the British National Bibliography and the National Lending Library for Science and Technology should be combined into a single organization to be called the British Library. This federation of institutions should be powerful enough to meet all of the objectives at which the government is aiming—comprehensive collection, availability for reference, lending, photocopying, cataloguing and the like. The questions now to be asked are questions of practice and not principle. Can the British Library look forward to the kinds of funds with which it may be expected to function well, and in particular will there be funds available to repair the long-standing and scandalous lack of buildings for some of the more important libraries? Will the British Library be competent to carry out the central bibliographical service which has hitherto fallen awkwardly between several institutions? Will the new organization be able to operate efficiently enough to provide the service which scholars of all kinds need at a price which governments and taxpayers can afford?

On the face of things, the government intends to be generous. The White Paper does, for example, acknowledge that the cost of building a new house for the reference libraries, on seven acres opposite the present British Museum in Bloomsbury, will be £36 million spread over a period of thirteen years. The difficulty is, of course, that at least thirteen years have gone by since it was first openly acknowledged that the National Reference Library of Science and Invention (then the Library of the Patent Office) has been outrageously neglected. At present, as a result of administrative improvisation, the library is divided between two sites several miles apart. Moreover, it seems quite plain that the library would equip itself with a wider range of materials drawn from a wider geographical area if only it had the funds. How soon will this neglected library be rehoused? And when it has a decent roof over its head, will it have the money with which to buy the books it needs? Because the National Reference Library for Science and Invention has been a pensioner of the British Museum at least to the extent that it has enjoyed the privilege of a free copy of each new book to appear in the United Kingdom, it has found itself lumped in with libraries which have a much smaller need to acquire material from abroad. But if this scientific library is the most seriously deprived of the national libraries, it is also plain that the library of the British Museum itself is under severe pressure. It is, after all, two years now since the library found it necessary to restrict the issue of readers' tickets to scholars from abroad. The danger in the government's plan is that by linking the future of these two

libraries to a gigantic rebuilding scheme, penury will be perpetuated. Is it not feasible to get the job done in less than thirteen years from the point at which the government chooses to put its proposals to Parliament? This is a point on which to press Lord Eccles, the minister in charge, and, until the change of government, ironically the chairman of the Trustees of the British Museum.

On the relationship between the constituents of the new British Library and other libraries, the White Paper is reticent to the point of coyness. To be sure, it does dissent from the Dainton committee's view that the Science Library of the Science Museum in South Kensington should be transferred to Imperial College, lock, stock and barrel. Instead, at least a part of the library will become a national library specializing in history of science. This is sensible, not merely because of the need for such an institution but also because there is a danger that the other busier institutions in the new complex will neglect this important field. Here again, it is a crucial question to know whether the library at South Kensington will have the funds necessary to acquire the ingredients of a collection sufficiently good to justify the government's claim that the new arrangements will provide a "system without rival". The relationship between the British Library and the university libraries may be in the long run even more important and, even though the government has the example of the Dainton committee to excuse its running away from this problem, the fact remains that some better machinery for coordination of library policy including the universities would be a national asset. It is true, of course, that university libraries quite properly develop lines of interest which are relevant to local scholarship, and there are obvious administrative difficulties stemming from the way in which university libraries must be financed by funds drawn from the University Grants Committee. It is also fair to say that university libraries are beginning to collaborate among themselves on issues such as the coordination of policy and even the acquisition of material from abroad—the committee under Dr Thomas Parry which reported to the University Grants Committee in 1967 has been a helpful spur. Yet much more needs to be said about the ways in which the new British Library can

Postal Strike

DURING the postal strike in the United Kingdom, correspondence for *Nature* from overseas may be sent either to the Washington office, the address of which is shown on the contents page, or c/o Messrs Gill and Macmillan, 2 Belvedere Place, Dublin 1, Eire. Advertisements for T. G. Scott and Son Ltd may also be sent to these offices (or by telephone to 01 242 6264). *Nature's* London office can also be contacted by Telex, 262 024.

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serve university libraries than has been hinted in the White Paper.

The organization of the British Library is a crucial matter. The government's intention is that the library should be independent of the government—with the constitution of a public corporation—but that it should be financed from the Department of Education and Science. Quite apart from the difficulty of persuading those now in charge of the constituent libraries to sink their futures in a common organization, there will be great difficulty in planning with sufficient care the development of bibliographical services for the United Kingdom as a whole—one of the most serious defects of the present system. The Dainton committee provided ample evidence that the important British libraries are seriously deficient in data

processing equipment. It is also clear that the catalogues the libraries maintain leave a great deal to be desired. The Dainton committee, for example, found the catalogues upon which loans among libraries are based are incomplete and that the National Central Library is in part responsible. What the board of the British Library will have to decide right from the beginning is how to plan for the use of computing machinery for keeping a catalogue, for sorting it and for using it as a means of identifying library material. It will be necessary, in the process, to develop means of matching this system with other systems now being developed, in the United States and elsewhere. It will take courage as well as imagination to make sure that this work is mounted early in the history of the new library.

Pricing Away the Market

THE amiable Mr Tom Jackson, the General Secretary of the British Post Office Workers Union, seems bent on demonstrating to his members and their employers that the mails are quickly becoming an anachronism. In the past few years, not merely in Britain but in the United States, the mails have been under as severe an economic pressure as any other labour intensive industry. The offer of an 8 per cent increase of salary which Mr Jackson has rejected would alone add close on £20 million a year to the cost of operating the postal service, itself nearly £400 million a year. Mr Jackson wants twice as much—in other words, a 15 per cent increase of salary or something like a 10 per cent increase in the cost of operating the postal service. This is a measure of how costs must necessarily increase when more than 70 per cent of the budget of the postal services is spent on wages and salaries. Inevitably, any settlement with the Post Office Workers Union, even the modest one which Mr Jackson scorns, will mean that there must be higher prices to the public, for one of the difficulties now apparent is that so long as the Post Office pretends to offer a nationwide service, with fast delivery to all points for the same charge, there is very little scope for reducing the overhead costs of the postal services. Yet the amount of traffic is steadily declining. In Britain, the postage of letters has been declining since 1968. With parcels, the British Post Office has been losing business since 1966. Any price increases now are bound to diminish still further the willingness of potential customers to use the mails. Even the preparations being made earlier this week to carry on without the Post Office will have tempted many potential customers to make permanent alternative arrangements, while the strike which began on Wednesday, however short its duration, may well have put an end to the national Giro system. Who wants a bank that closes down?

In circumstances like these, the time has come—indeed, it came a long time ago—when the British Government and the Post Office Corporation should take a close look at the function of the mails in an industrial and highly developed country such as Britain. If anybody needs a crib, he can find one in the report of the Commission on Postal Organization published in the United States in June 1968. There, where more than 150,000 million pieces of mail are delivered every year, the outstanding problem is that the postal services depend on what is in effect a

subsidy of close on 20 per cent from the Federal budget. Yet productivity is growing much less quickly than in other industries—output per man measured as pieces of mail handled each year has increased merely by 0.23 per cent a year in the past decade, chiefly because of the failure to use better methods of handling and of management. Surveys have, however, shown that even with the cheap rate for what is called junk mail, something like 40 per cent of all items posted consists of business correspondence—cheques, invoices and the like. Both the government and Mr Jackson should acknowledge that business of this kind is more easily converted to other methods of communication than most other kinds of mail.

Even in Britain, once commercial banks are properly provided with computers, a good deal of this traffic in commercial paper can be made electronically. In due course, no doubt, when the brave new world comes true, the telex terminal in every household will provide a more convenient way of doing the same job. But this is only the obvious beginning. Already there are signs that the distribution of pieces of paper marked with ink in various ways does not always require physical transport from origin to destination. One obvious illustration, already a reality, can be seen in the way that lithographic printing has made it possible for identical books or magazines to be produced simultaneously and economically in several places—the economies of scale in a single printing are not always sufficient to offset the cost of distribution. But in due course there will be a photocopying machine on every television set and then, no doubt, the mechanics of distributing paper as Post Offices have done for the past century will be radically transformed. How then should the British government, with or without the connivance of Mr Jackson, arrange to live with an industry in which costs must inevitably rise and traffic diminish? The first thing to decide is that nobody can hope to profit from an attempt to recapture the euphoria of the 1840s, when the penny post seemed for a time to make Victorian England safe for centuries. The economic cost of delivering a letter in Britain is probably now more like ten times as much and rising rapidly.

The best service the government could now perform for the postal services would be to devise some relationship between the cost of handling each item of mail and the charge made for doing so. Why not, for example,

an extra charge for the delivery of mail to people's houses or business offices? The foolish insistence on sending a man in uniform to each of the 17 million households every day is one of the causes of the insolvency of the postal service. And are there not circumstances in which some geographical spread of charges would in the long run be economic? Would it not even make sense to split up the postal services on a regional basis? The cost of carrying mails from the south of England to the north of Scotland is, after all, by no means negligible. The Select Committee on Nationalized Industries uncovered the significant information that the cost of handling an item of rural mail is something like 40 per cent greater than that of handling a piece of mail collected in the city. Given that telephones have now been in use for close on a century, has not the time arrived when the Post Office should ask its country customers to pay a little extra not merely for the sake of their city cousins but rather so that the postal services shall survive in some form or another? And is there not a case for qualifying the monopoly which the Post Office enjoys on the delivery of mail, which has on occasion prevented other nationalized industries such as the electricity and gas utilities from distributing their own mail? Nobody would ask for changes that would rob the Post Office of its bread-and-butter, but there are already provisions in the Post Office Act for shading the traditional monopoly of the Post Office Corporation. It would be sensible now if the Post Office were to set about hiving off on a commercial basis those parts of its traffic which are most embarrassing financially.

Better management may help to keep the wolves at bay, but to the extent that the government and the Post Office Corporation have the public interest at heart, there is also a strong case for asking that they should between them encourage the development of those devices that will in the long run help to take the strain off the postal services. Quite simple devices could do a lot to help. At present, for example, it seems most probable that the capital equipment of the telephone network is much less fully used than it might be, partly because of the lack of switching equipment and partly because the tariffs at present levied by the Post Office are not designed to encourage traffic. If in due course the rural users of the postal services are to be given an economic alternative, this is where the Post Office should put its efforts. If the fixed element of the rental charge which telephone subscribers pay is actually excluded, the average telephone call costs the customer about the same as a first class letter. Even with the rental charge included, the cost is only twice as high. But everybody knows that the telephone is at once less convenient and less cheap than other devices, telex for example. So is not the government's duty to require first of all that postal carriage should match economic costs so that existing telecommunications services can compete effectively with the methods of the 1840s, and then that large sums of money should be spent on the development of the radically different systems that the 1980s will require? This is what Mr Jackson's understandable intransigence has demonstrated.

In the recent history of the British Post Office, there is nothing to suggest that these opportunities will be seized upon. When the Post Office was a government department, it was renowned for its conservatism. Theoretically, making it into a public corporation should have invested it with a more vigorous competitive spirit. In the event,

however, the public corporation turns out to have all the disadvantages of a privately owned company and few of the advantages. Lord Hall, the last chairman—no successor has been named—enjoyed the apparently compliant support of his colleagues on the corporation but was sacked by the minister as if he had been an errand office boy. By the same test, the corporation is able to put its hand on the very large sums of money needed to keep the telecommunications network extending over the country without having to compete for these in public, simply because nobody wishes to see the telecommunications network languish. Yet the corporation's stake in research and development, let alone systems planning for the future, is much less expert than it should be. Commercial organizations in the strict sense would never dare to make the assumptions about the affections of their customers which even the devolved Post Office Corporation takes for granted. What the strike has done is to give the Minister of Posts and Telecommunications even greater authority over the Post Office than his predecessor dared to exercise. Is it, perhaps, that the British government is preparing the way for a different recipe for devolution? This is another of the hornets' nests that Mr Jackson may have disturbed.

100 Years Ago



The Primary Colours

I HAVE been greatly interested in reading Mr. Strutt's curious experiments on colour in the last number of *NATURE*. I am glad to see that he is able to assume as proved the theory that green and not yellow is the middle primary. The true position of green is well illustrated in Mr. W. Benson's "*Principles of the Science of Colour*," both by argument and by diagrams. There is, however, one piece of evidence which seems to me conclusive as against yellow, but which I have not seen noticed.

When a solid body is gradually heated to incandescence, the light given out is first red, then orange, afterwards yellow, and finally white. If yellow were a primary, it would be impossible for it to appear in this series, which is formed upon the basis of the first primary, red, by successive additions of more and more rapid vibrations. Every colour in the series except the first must be a compound. If the heat is not sufficient to generate the most rapid vibrations which the eye can appreciate, white light is not given off at all, the series terminates with the yellow. The light of a glowing coal, without flame, in an ordinary fire, rarely passes beyond the yellow stage, and such light yields to the prism abundance of red and green rays, but scarcely a trace of blue or violet.

But, if red is the first primary, and green the second, which is the third? Shall blue still sit upon the throne on which Newton placed him, when his brother yellow is deposed? I think his position has become extremely precarious, and that he would be wise to abdicate with dignity before he is ignominiously turned out as a usurper.

If the kingdom of light is really divided into three principalities, is not violet the rightful heir to the third throne? Violet is said to be a mixture of blue and red. But how should red make its appearance at the wrong end of the spectrum? If it has no definite limit, but gradually thins out from its own place to the other extremity of the spectrum, then the whole of the other colours must be more or less affected by it, and red must be the only true primary among them. If it is said that the red in violet is clearly recognised by the eye, I think it may be answered that this is only because we have been taught to think of it as a compound, and that we might just as truly say that we can see yellow in green or orange in red.

Leicester, Jan. 21

FREDK. T. MOTT

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OLD WORLD

HIGHER EDUCATION

Recipe for Change

by our Education Correspondent

A GROUP of individuals from higher education, government and industry has put forward a set of suggestions for revamping higher education which are far reaching and yet retain some of the flavour of the traditional British university system. Called the Higher Education Policy Group, and convened by Professor W. R. Niblett of the University of London Institute of Education, the group has been discussing the future development of higher education in Britain for the past 18 months. The suggestions offered, although not spelled out in detail, include a flexible system which would enable a student to qualify for a degree in two years, or an honours degree in three, a unified admissions procedure for all institutions of higher education, ease of transfer between institutions and more autonomy for the public sector.

This statement (available from the University of London Institute of Education, 1s 6d) is the latest in a series of recent reports and suggestions for revamping higher education, all of which have sprung from acknowledgment of the fact that the population of the universities is likely to grow faster than their finances, and some way of expanding higher education within strict financial limits must therefore be found. But the Higher Education Policy Group also believes that there are strong educational arguments for changing the present pattern of courses and the administrative structure of higher education. The group is also the first to admit that it offers no panacea for the problems.

The group's chief suggestion is that although an honours degree should be awarded only on completion of three years full-time study or its part-time equivalent, students should be able to alternate between full-time and part-time study. And the policy group also floats the idea that students should be able, at their own choice, to leave higher education after two years, taking with them a degree. This implies that universities should offer part-time courses, and that students should be able to transfer from one institution to another. The policy group is therefore aiming for a unitary system of higher education, at least as far as courses are concerned.

A unified admissions procedure for all institutions of higher education and a system of course credits are necessary corollaries to such a suggestion. Although the policy group discusses the first in some detail, it gives only passing reference to the second. It suggests that a first

step in rationalizing admissions systems would be for the polytechnics and other similar institutions to agree on a single common application form, and for the separate admissions procedures for universities and colleges of education to be merged. Unless some common admissions procedure along these lines is accepted, the present system, in which students are forced to choose between a university or some other establishment, will continue to ensure that polytechnics attract applications chiefly from students unable to gain admission to the universities.

Recognizing that there will be a considerable diversity of institutions, at least for the next ten years, the Higher Education Policy Group suggests that administrative differences should not be allowed to distort their underlying community of purpose. The total cost of teaching a first degree student on a given type of course in a given subject should, for example, be broadly similar throughout the whole sector, and there should be more sharing of expensive academic equipment between neighbouring institutions. The policy group warns that unless institutions of higher education take active steps to achieve closer coordination, "they are likely to have a less acceptable form of rationalization imposed upon them".

As far as the public sector itself is concerned, the policy group believes that some local authority representation in the control of finances for the polytechnics is desirable, but the group suggests that a central body should be established at national level for approving capital expenditure on higher education. The Department of Education and Science should determine the overall capital expenditure on the public sector of higher education, and the new body should allocate projects within the total budget.

How would the colleges of education fit into the proposed scheme? The Higher Education Policy Group puts forward three alternative suggestions. Some of the larger colleges might develop into degree-giving institutions, offering a wide range of choices in sciences and liberal arts; others might become federated or integrated with universities or polytechnics, and the remainder might concentrate on in-service training for a variety of professions.

Will these suggestions result in much saving in unit costs? The policy group realistically suggests that only about 10 per cent of the students will opt to leave higher education after two years, and the saving in costs would therefore only be of the order of £35 million in 1981. The group also believes, however, that a reduction of about 15 per cent in the staff/student ratio in higher education as a whole should be tolerable over a ten year period of expansion, and

this would save some £60 million in 1981.

Such savings would be unlikely to satisfy those who believe that the higher education system already takes an excessive share of the gross national product, and the Higher Education Policy Group is therefore forced to suggest that a partial student loans scheme should be considered. Although the group takes pains to point out that it is in principle opposed to any financial arrangements which may act as a deterrent to entry to higher education, it suggests a scheme in which maintenance grants would continue at their present level, so that their real value would be eroded by inflation, and a student would be able to take out a loan to be paid back at a prearranged percentage of his future income. If the average sum accepted by each student were £100 a year after 1971, the policy group calculates that repayment would be about £30 million by 1981 and more than £100 million in 1991.

Even with savings of this order, however, the Higher Education Policy Group's suggestions would still entail an increasing percentage of the gross national product being devoted to higher education. It would therefore be necessary to look towards savings in research costs, and to the whole relationship between teaching and research. There is also good reason to take a look at the research establishments of the research councils to see whether there is scope for their integration with the universities. The Higher Education Policy Group has conspicuously left aside such considerations. Nevertheless, its suggestions for reshaping courses form a compromise between the radical proposals put forward by Professor Pippard and those who believe that the system should be allowed to develop much along its present lines.

The following people signed the statement: S. L. Bragg, Michael Brock, G. S. Brosan, Colin Crouch, Frederick S. Dainton, Goronwy Daniel, Bernard de Bunsen, Brian H. Flowers, R. C. Griffiths, P. D. Hall, F. R. Hornby, H. G. Judge, P. R. G. Layard, Norman Lindop, William Mansfield Cooper, A. W. Merrison, D. E. Mumford, W. R. Niblett, Beryl Paston Brown, James F. Porter, Marjorie E. Reeves, and Bryan Thwaites.

BIOLOGY TEACHING

Showing the Way

SOME far-reaching changes are on the way in the teaching of biology at British universities. This was made clear when members of the Institute of Biology met last week at the Royal Society, London, to discuss the problems they face in an age of university expansion and financial

stricture. To compare the ways in which some of these problems may be overcome, participants took with them an impressive collection of visual aids and equipment.

Professor H. E. Street (University of Leicester) set the scene with an account of the difficulties now inherent in teaching biology in universities. The diversity of backgrounds of the students, with some lacking O-level physics and mathematics or A-level biology, will probably increase as universities expand. The task of bringing such students to the same level of knowledge is formidable, and Street said that, at Leicester, efforts so far have been fraught with difficulties. His department, for example, has tackled the problem of the lack of, or aversion to, mathematics by establishing a course known as "quantitative biology". But there is disagreement about the status of this course, about its content and about the time which should be devoted to it.

One of Street's most strongly expressed points was that students are increasingly questioning the relevance of almost everything they have to do. He emphasized that it is vital to persuade the student who intends to become a microbial geneticist that it is valuable to study the morphology of the leaf, remote as it may be from his chosen career, because it will help him to become a better biologist. For situations such as this, Street felt that interdisciplinary teaching is essential.

One outcome of the concern for problems of this sort has been the Inter-University Biology Teaching Project. Financed by a grant of £80,000 from the Nuffield Foundation, the Universities of London, Glasgow, Birmingham, Sussex and Bath have worked out various courses, making use of programmed texts, films, videotapes and other visual aids. The courses, described by Professor O. E. Lowenstein (University of Birmingham), are mostly either bridging courses, designed to make good deficiencies in knowledge, or service courses for students who wish to be introduced to a subject, not necessarily with a view to later specialization.

AGRICULTURE

Prior's Priority

ONE of the first signs of the severity with which the government regards its resolve to cut public spending came last week with the announcement by the Minister of Agriculture, Mr James Prior, that many of the services which the ministry provides for farmers are to be discontinued. These measures, which should save the department some £15 million a year, will be spread over three years and may mean that as many as 1,500

(10 per cent) Ministry of Agriculture civil servants will lose their jobs, although Mr Prior believes that much of this staff saving would come through natural wastage.

The greater part of the economies which Mr Prior proposes in his White Paper (Cmd 4564) lies in the cancellation of a number of the services to farmers which in the past have been provided free. The County Agricultural Executive Committee system will be brought to an end together with the Agricultural Marketing Facilities Committees and the local Wheat Committees. Some thirty other advisory bodies including the North Pennines Rural Development Board and the Council on Rabbits and other Land Pests will similarly be axed. The plan is that farmers who are progressive and interested in improving their farms and production should seek out the ministry's help, or alternatively obtain advice and help from private enterprises.

This represents a clear change in the government's attitude towards the farmer. Whereas in the past the advisory committees have come to the farmer, it will soon be the other way round. The farmer is being asked to show, in Mr Prior's words, "individual initiative and self-reliance". It seems clear that the cuts will strike hardest against the small and the inefficient farmer, whereas the wealthy or cooperative farmer will not suffer to the same extent.

ELECTRONICS

Siemens's UK Expansion

THE formation of an electronics division within Siemens (UK) Ltd is the latest development in the re-establishment of the well known German electrical firm in Britain after two setbacks during the world wars. A Siemens subsidiary was first founded in Britain in 1858 only 10 years after the formation of the parent company, Siemens AG, in Berlin. Siemens AG has now grown to be the largest electrical concern in Europe after Philips with sales which reached £1,400 million in 1969-70.

For 20 years after the Second World War, marketing of Siemens products in the UK was entrusted to various agencies. From 1965, however, Siemens (UK) Ltd was able to resume the firm's operations under its own name but since then the company's chief role has been that of an importer although some assembly and manufacturing work is now carried out at a factory in Cheshire. The new electronics division will be situated at Croydon, Surrey, and will market a range of Siemens products which broadly reflect manufacturing divisions within the parent company.

Parliament in Britain

Aldermaston

THE proportion of the total expenditure attributable to civil work at the Atomic Weapons Research Establishment, Aldermaston, has increased from 17 per cent in 1967-68 to 19 per cent in 1969-70. This information was given by Mr Nicholas Ridley, Undersecretary of State, Department of Trade and Industry, in reply to a question from Mr Albert Booth. But Mr Booth suggested that there is still an urgent need to transfer a higher proportion of research and development capacity in Britain to civil research.

In reply to a later question from Mr R. Carter, Mr Ridley announced that government support for the Aldermaston project for the application of computers to engineering is being scaled down. The need for such support is no longer apparent, he said, and the future of the project must depend on the extent of its financial support from industry. Although the project has done valuable work in the past, Mr Ridley said there is no reason why industry should not pay for it, whether the work is carried out in the public or the private sector. (Oral answers, January 18.)

Atomic Energy Authority

SIR JOHN EDEN, Minister for Industry, gave an assurance during the committee stage of the Atomic Energy Authority Bill that employees transferred from the AEA to either of the proposed companies—British Nuclear Fuels Ltd and the Radiochemical Centre Ltd—will be employed on the same conditions as they enjoyed immediately before transfer. Sir John Eden rejected, however, the suggestion from the staff of the Atomic Energy Authority that there should be a permanent link between the terms and conditions existing in the proposed companies and those existing in the Atomic Energy Authority at any time in the future. He said that after the interim period, adequate negotiating machinery would be established in the new companies. He said, however, that a temporary link between the pay scales in the new companies and in the Atomic Energy Authority would be maintained.

An amendment to the bill, moved by Mr Airey Neave, which would enable the Secretary of State for Trade and Industry to set up subsidiaries of the proposed companies was withdrawn after Mr Nicholas Ridley had given an assurance that British Nuclear Fuels Ltd would be able to hold interests in other companies and to enter into any arrangements with other companies. The company can therefore set up operational subsidiaries in selected areas on its own initiative, and such a move would not be the responsibility of the Secretary of State. (Debate, January 15.)

NEW WORLD

Where to Get your PhD

by our Washington Correspondent

HARVARD has the most distinguished molecular biology faculty of any university in the United States, but CalTech is the best place to get a doctorate in the subject. The University of Iowa fields one of the most distinguished pharmacology departments, but its population biology course for graduate students is only adequate to good. Ninety-three per cent of a group of scientists consider their colleagues at Harvard constitute the most distinguished chemistry faculty in the nation; 10 per cent aver that Harvard's civil engineers are less than adequate.

This and much other comparative detail is offered in a new shopper's guide* to graduate departments issued this month by the American Council on Education, an association of colleges and educational institutions. The purpose of the survey is primarily to assist prospective graduate students with information about the professional standing and effectiveness of various graduate programmes; another motive is to indicate to academic administrators which departments are giving value for money and which are lagging behind.

The basis of the survey is a questionnaire mailed in spring 1969 and returned by some 6,000 faculty members in 36 disciplines (including 10 biological sciences and 5 physical sciences). Each participant was asked to rate the institutions with graduate faculties in his field according to a five point scale ranging from distinguished (5 points) down to strong, good, adequate and marginal (1 point). Separate questions asked raters to assess the effectiveness of the doctoral programme and the change in the quality of the programme occurring over the past 5 years. To be included in the questionnaire an institution had to have awarded at least 100 doctorates in two or more disciplines in the past 10 years, and at least one doctorate in the discipline under assessment. As a precaution against generation gaps, the judges were chosen to include chairmen of departments, "senior scholars" and "junior scholars" (the latter having held a doctorate for less than ten years) but in fact the assessments of all three groups were highly correlated one with another.

The design of the study essentially followed that of a survey conducted by

the council in 1964 but the presentation differs in that the results have been deliberately obfuscated so as to "de-emphasize pecking-order relationships" and avoid the bolstering or deflation of egos. Thus the council has coyly suppressed the precise scores gained by its individual members, reporting only the broad class to which each graduate faculty is assigned. The four classes, labelled "distinguished" and "strong", "good", "adequate plus" and "all other", correspond to average score ranges of (3.0-5.0), (2.5-2.9), (2.0-2.4) and (0-1.9) respectively. Within the first class only (distinguished and strong), graduate faculties are listed in rank order of their ratings. In the second two classes they are listed alphabetically and for the bottom class (those judged by raters as "adequate" or "marginal") the council names no names.

The survey leaves readers to compile their own lists of the top ten universities; one criterion, that of the number of appearances within the top five places in all disciplines, gives the following

rank order: Berkeley (among top 5 in 32 disciplines), Harvard (27 disciplines), Stanford (16), Chicago (14), Yale (13), Michigan (12), MIT (12), Princeton (12), CalTech (11), Wisconsin (8). Considering only the fifteen pure sciences, the rank order (by the same criterion) is Harvard and Berkeley (13 each), Stanford and CalTech (8 each), MIT (6), Rockefeller (5), Chicago, Princeton and Wisconsin (4 each), Yale (3) and Michigan, Illinois and Cornell (2 each). In the biological sciences Harvard's graduate faculties are rated top in the disciplines of molecular biology, biochemistry, physiology and zoology, whilst Berkeley takes the palm in botany, entomology and zoology. Other firsts are CalTech in developmental biology, Rockefeller in microbiology and physiology (tied with Harvard) and Iowa City, Minnesota, Stanford and Yale, judged equal first in pharmacology. As for the physical sciences CalTech is top of the poll for astronomy, geology and physics (equal first with Berkeley and Harvard). Harvard is best for chemistry, Harvard

Table 1. Rank order of institutions in which the quality of the graduate faculty was rated either "distinguished" or "strong". * means rank shared with another institution; † means 1969 score is better than 1964; italics means institution not included in 1964 survey of this discipline.

Molecular Biology			
1	Harvard	3	California, Berkeley
2*	California, Berkeley	4	Chicago
2*	CalTech	5	Harvard
4	Stanford	6	Wisconsin
5*	MIT	7	Arizona
5*	Rockefeller	8	Cornell
7	Wisconsin	9*	MIT
8	Johns Hopkins	9*	Maryland
9*	California, San Diego	11*	California, Los Angeles
9*	Chicago	11*	Columbia †
9*	Yale	11*	Texas
12	Princeton	14	Michigan
13	Brandeis	15*	Washington (Seattle)
13*	California, Los Angeles	15*	Yale
13*	Cornell	Geology	
13*	Illinois	1	CalTech
13*	Washington (Seattle)	2*	California, Berkeley
18	Purdue	2*	Harvard
19*	Oregon	4*	MIT
19*	Pennsylvania	4*	Stanford
19*	Yeshiva	4*	Yale
22*	Duke	7*	California, Los Angeles
22*	Michigan	7*	Princeton
22*	Washington (St Louis)	9	Columbia
25	Columbia	10*	Chicago
26*	California, Davis	10*	Penn State
26*	Indiana	10*	Texas
26*	NYU	13*	California, San Diego
29*	Case Western Reserve	13*	Johns Hopkins
29*	Michigan State	13*	Michigan
29*	Minnesota	13*	Northwestern
29*	Texas	13*	Wisconsin
Astronomy			
1	CalTech	18	Minnesota
2	Princeton	19*	Illinois
		19*	Rice
		21	Brown

* *A Rating of Graduate Programs* by Kenneth D. Roose and Charles J. Andersen. American Council on Education, 1 Dupont Circle, Washington, DC 20036. \$4.00.

and Berkeley are best for mathematics.

From the item on the questionnaire concerning the change in the quality of graduate teaching the authors of the survey are able to report a notable improvement—out of 1,600 graduate programmes three-quarters were considered to have changed for the better and 80 per cent were considered to be adequate or better, compared with only 70 per cent at the time of the last survey in 1964. Against this average improvement, there has been a general drop in the proportion (though not in the absolute numbers) of programmes considered strong and distinguished, except in four disciplines (geography, psychology, sociology and entomology) where the proportion of first class departments has increased.

The authors of the survey draw three general conclusions from the data they have gathered, one trite and two important. First, it would be a pity if institutions were led by the study to improve their graduate programmes at the expense of undergraduate studies. Second, there appears to be some duplication of graduate programmes, particularly by the public institutions within a given state. Far too often two state universities vie with each other in identical fields, or even across the board, with the result that resources are spread too thinly, instead of agreeing on some rational division of labour. "This criticism," the authors remark, "appears especially pertinent to those institutions that have been less well established in graduate fields." On the other hand, those states where several universities offer high quality programmes in the same discipline should consider whether they are making the best use of the increasingly scarce resources for higher education. Because of the likely shortage of funds, academic administrators should probably abandon hope of raising their below-par programmes up to scratch and should instead decide to drop them altogether.

The third conclusion of the survey is that the number of graduates being churned out by the university system may be excessive. For example, the projected output of PhDs for 1980–81 is computed at 50,900, whereas the total demand for new PhDs in the teaching system in the same year is only 7,400. This somewhat imprecise foreboding should be set against the admittedly optimistic projections recently issued by the Bureau of Labor Statistics, in which a 54 per cent increase is foreseen in the demand of private industry for PhD scientists and engineers in 1980 compared with demand in 1968. The projected demand for 1980 totals 55,000 PhDs (compared with 35,800 employed in 1968), of which 20,100 are engineers, 1,300 mathematicians, 29,000 physical scientists and 4,100 life scientists.

CHEMICAL WARFARE

Herbicide Commission reports Extensive Damage

by our Washington Correspondent

THE military use of herbicides in South Vietnam has caused extensive and lasting damage to vegetation but the effects, if any, on human health are as yet unclear—that is the chief verdict (as reported in *Nature*, 229, 81; 1971) of the Herbicide Assessment Commission appointed by the American Association for the Advancement of Science. A detailed report of the commission's work is to be published later, but preliminary results, of which the following is an account, were announced at the annual meeting of the AAAS in Chicago last month.

The commission visited South Vietnam for a five week period in August and September last year after several months of advance planning. Five aspects of the herbicide programme were chosen for study: (i) the effects on human health, (ii) the effects of the defoliation programme on the mangrove swamps and (iii) on the tropical hardwood forests, (iv) the effects of the crop destruction programme and (v) the distribution of the herbicides and their residues and contaminants in food chains. Members of the commission were Matthew S. Meselson, professor of biology at Harvard University, who was appointed by the AAAS board in December 1969 to design the study, Arthur S. Westing, professor of botany at Windham College, Vermont, John D. Constable, professor of surgery at Harvard Medical School, and Robert E. Cook, a graduate student in ecology at Yale University.

Between 1962, when the herbicide programme got under way, and 1969, the last year for which full data are available, some 22,000 square kilometres, or nearly one seventh of the total land area of South Vietnam, have been sprayed, including an estimated 20,000 square kilometres of forest and 2,000 of cropland. As far as concerns human health, the most important of the three principal chemicals used in the herbicide formulations is 2,4,5-T (2,4,5-trichlorophenoxyacetic acid), which has been shown to produce birth defects in certain animals under some laboratory conditions. Also of interest is an impurity introduced in the manufacture of 2,4,5-T known as dioxin or 2,3,7,8-tetrachlorodibenzodioxin. Besides inducing birth defects in laboratory animals, dioxin is exceedingly toxic, its LD₅₀ for guinea-pigs being 0.0006 mg/kg. Since the chemical may persist in the environment and is soluble in fat, it may be concentrated as it moves up the food chain into human diet. According to rough calculations the Herbicide Assessment Commission believes it is "not impossible that significant amounts of dioxin are

entering the Vietnamese diet". The commission brought back samples of food and human tissue to test this possibility but has not yet been able to develop a suitably sensitive test for measuring the minute quantities of dioxin the samples may contain.

As an alternative approach to the question the commission examined South Vietnamese hospital records for any change in the pattern of births. An independent study carried out by the US Army and the South Vietnamese Ministry of Health, first made known on December 22 last year, surveyed hospital records over the last ten years and reported a slight downward trend in birth defects, concluding that the data "failed to show any influence of herbicides anywhere". The Herbicide Assessment Commission disagrees with both the conclusion and the data of the Army study. People living in the country would have been more heavily exposed to herbicides than those in Saigon, from which most of the data are taken. Subtracting the Saigon data, the Army study shows a decided upwards trend in the three kinds of birth defects (stillbirths, placental tumours and deformities). In fact probably less than 5 and perhaps less than 1 per cent of the South Vietnamese population has been directly exposed to herbicides, a factor which is unlikely to show up when the birth statistics of the country as a whole are considered, particularly since many of the exposed population are probably the montagnard people among whom births usually take place at home and do not reach the South Vietnamese medical records.

Rather than trying to survey the birth records over a large area, the commission concentrated its efforts on the province of Tay Ninh where the forests have been heavily sprayed and the inhabitants eat the fish in rivers draining out of the sprayed areas. Examining the original records in the Tay Ninh provincial hospital the commission noted 351 stillbirths in the years 1968 and 1969—the Army study, which did not have access to these records, reports only 208 stillbirths—making for a stillbirth rate of 58 per 1,000. During the same period the rate for the Tu Du hospital in Saigon was 28 per 1,000 and that of the entire country was 31.2 per 1,000. The commission notes that this type of evidence is "not sufficient to draw any firm aetiological conclusion".

Similarly indicative but inconclusive evidence comes from a study of common birth defects in the Saigon Children's Hospital. Total admissions have remained constant at about 600 cases a year from 1964 to 1968 but there has been a

"very striking increase" in the incidence of spina bifida (less than 5 cases a year until 1966, 13 cases in 1967, 12 in 1968) and of cleft palate (over the five year period the number of cases were 5, 2, 12, 23, 13). The other anomalies treated in the hospital, the only special children's hospital in Vietnam, exhibited no demonstrably significant change over the period. The commission notes that the increased incidence of spina bifida and cleft palate could be the result of improved diagnosis, but chooses to describe them as unexplained. Despite contrary reports in the Vietnamese press, the commission found no evidence for the occurrence in recent years of rare and striking birth defects such as the absence of limbs caused by thalidomide, but notes that much of the population directly exposed to herbicides was not available for study.

The impact of the herbicides on the forests of South Vietnam is not as difficult to assess. For example the mangrove forests, which cover much of the coastal area of the Mekong Delta region, have proved for unknown reasons to be particularly sensitive to herbicides, essentially all vegetation being killed by a single spraying. By the end of 1967 about a third of the country's 2,800 square kilometres of mangrove forest had been sprayed and the proportion is now close to one half. According to Arthur Westing, the botanist on the commission, "the scene of a spray attack is a weird and desolate one.... Herbicidal attack appears to prevent the reestablishment of any new plant community... for at least six years, and clear evidence of recolonization has not been observed anywhere by us or others". A rich variety of fish and crustaceans live in the channels of a mangrove forest or offshore, supported directly or indirectly by the nutrients flushed out of the forest. The commission has no proof that these populations have dwindled since the death of the mangroves but believes this is likely to have happened. Marine erosion is another possible consequence the commission was unable to assess. Situated at the cutting edge between land and sea, the mangroves may serve the purpose of stabilizing the shoreline against the action of winds and tides. Although no serious evidence of marine erosion has yet appeared, this is a threat that could materialize as the dead roots rot away. There has been no major typhoon since the herbicides were sprayed.

In the tropical hardwood forests, which occupy about 100,000 square kilometres of South Vietnam, the effect of herbicides has been less devastating but equally hard to assess in the long term. Some 20,000 square kilometres are thought to have been sprayed, a quarter of them more than once. Westing estimates that at least one out of every 8 or 10 trees is killed by a single spraying, rising to anywhere from 50 to 80 per cent or higher in the areas sprayed more than

once. The immediate consequence of the defoliation is the loss of the nutrients contained in the fallen leaves. Because of rapid decomposition and heavy rainfall, the soil of South Vietnam's tropical forests cannot retain any significant fraction of the nutrients, most of which are lost to the system.

Two important conclusions reached by the commission about the crop destruction programme are, first, that it is probably ineffective in its stated objective of destroying crops intended for enemy consumption and, second, that its brunt has been borne by the montagnard tribespeople of the central highlands of South Vietnam. The commission also concludes that the precautions taken to avoid destroying crops grown by civilians for their own use have been a failure. Some 2,000 square kilometres of cropland has been sprayed since the programme began, thought to represent enough food to feed 600,000 people for a year. The commission believes that nearly all this food would have been consumed by civilian populations. The crop destruction programme has been largely confined to the food scarce central highlands, most of whose one million inhabitants are montagnards. The commission's verdict on the usefulness of the crop destruction programme in denying food to the enemy confirms that reached by several studies conducted for the Army. A survey completed by the Rand Corporation in 1967 concluded that only 5 per cent of enemy prisoners and defectors had depended on locally grown crops. A similar review last year inferred that less than one per cent of enemy soldiers grew their own crops although many more depended on civilian crops indirectly.

Other relevant evidence, concerning the persistence in the environment of the contaminant dioxin, was presented by Fred Tschirley of the Department of Agriculture. In experiments yet to be published he finds that dioxin lasts for a long time in soil, regardless of the microbiological activity; after 160 days some 95 per cent of the chemical could be recovered. But only a small proportion—a maximum of 0.16 per cent—is picked up by plants, the amount reaching a peak after 16 days and then declining. In sunlight dioxin is degraded quite rapidly but the important point of whether the chemical accumulates in fatty tissues is not yet known.

Finally, the commission noted that it was not directed to assess the military usefulness or desirability of herbicides but that this was a matter that could be objectively and quantitatively studied. Such a study is now unlikely to be made, for the Secretary of Defense announced on December 26, four days before the Herbicide Assessment Commission made its findings public, that there would be an "orderly yet rapid phase-out" of herbicide operations.

SALARIES

Scientists Earn More

THE slackening demand for scientists does not seem to have driven down their price on the market, according to data gathered by the National Register in spring 1970. The median salary reported by the 313,000 scientists answering the survey was \$15,000, an increase of \$1,800 (15 per cent) on the figure for 1968. Over the same period unemployment, though still small, has nearly doubled, from 2,800 (0.9 per cent of the registrants) to 4,900 or 1.6 per cent.

The range of salaries indicated by the survey extends from \$24,500 or more in the highest decile to \$9,500 or less in the lowest decile. Statisticians are the best paid discipline, earning a median salary of \$16,900, followed by computer programmers (median \$16,500) and economists (median \$16,300). Ranked by type of occupation, self-employed scientists gain the most just desert for their labours (median salary \$20,000); next are those employed in industry (\$16,700), by non-profit organizations (\$16,400), and in educational institutions (\$15,500 on a calendar year basis). More than a third of the registrants were engaged in some form of research—14 per cent in basic research, 13 per cent in applied research and 10 per cent in the management of the research and development.

The National Register covers about four-fifths of all doctorate scientists in the United States (including physical, biological and social scientists) and two-thirds of all qualified scientists. Full results of the survey, now in the press, will be published by the US Government Printing Office early this year (*Reviews of Data on Science Resources No. 19, Salaries and Selected Characteristics of US Scientists 1970*).

Washington Newsletter

A TWICE monthly newsletter specializing in relations between science and government in Washington is to start publication in February. Called *Science and Government Report*, the newsletter will be published by Daniel S. Greenberg, a former member of the news and comment staff of *Science* since 1961 and head of it for much of that time. Greenberg, who left *Science* last month, has recently spent two years based in London as the magazine's foreign news editor. The newsletter is intended for scientists, research and academic administrators and industrial research executives. Subscriptions are \$25 a year for US residents and \$35 for those with addresses outside the United States.

NEWS AND VIEWS

Hunting for Molecules among the Stars

INTERSTELLAR molecules seem to be starting the new year in a continued blaze of publicity, as the discovery of yet more complex molecules encourages increased attention from observers. In the last issue of *Astrophysical Journal* for 1970 (162, L203) the first six atom interstellar molecule was announced (see *Nature Physical Science*, 229, 36; 1971), and the first two issues of *Astrophysical Journal* for 1971 now report further developments in this fast growing area of astronomy.

Work carried out at the NRAO's Green Bank radio observatory in West Virginia has revealed the existence of microwave radiation characteristic of the cyanoacetylene molecule (HC_3N) in the emission from the galactic radio source Sgr B2. This discovery is particularly interesting because the molecules of cyanoacetylene are linear, with a configuration $\text{H}-\text{C}\equiv\text{C}-\text{C}\equiv\text{N}$; the spectra of such molecules are simplified because the electrons are paired off, leaving no net electronic angular momentum, and although there remains some splitting of the spectral lines caused by quadrupole interactions, this is smaller than the broadening of the lines arising from turbulence and the Doppler effect. B. E. Turner reports the presence of features attributed to ground state first transitions of this molecule, Doppler shifted by an amount corresponding to roughly 64 km s^{-1} , in good agreement with measurements of formaldehyde and hydroxyl features in the same source, all of which show velocities just over 60 km s^{-1} (*Astrophys. J.*, 163, L35; 1971). Although a search of other galactic sources containing molecules of OH, H_2CO , and HCN has failed to produce any more evidence for HC_3N , other reports recently published also add positively to the growing store of knowledge concerning interstellar molecules.

B. Zuckerman, of the University of Maryland, and J. A. Ball and C. A. Gottlieb, of Harvard College Observatory, have also used the 140 foot Green Bank telescope to good effect, and they provide more information about the microwave characteristics of formic acid, the simplest organic acid, in a letter in the second part of the same volume of *Astrophysical Journal* (163, L41; 1971). The source of this radiation is Sgr B2, which is rapidly becoming a star source for the hunters of complex molecules. A rather less definite observation of a line attributable to this acid has been made in the direction of Sgr A, but this is overshadowed by the Sgr B2 discovery, which again corresponds to a line Doppler shifted by a velocity close to 60 km s^{-1} .

The smaller telescopes at Green Bank are also providing valuable information. Using the 36 foot telescope, L. E. Snyder, of the University of Virginia, and David Buhl, of NRAO, have studied emission spectra from hydrogen cyanide molecules containing two different isotopes of carbon, H^{12}CN and H^{13}CN . This work shows how rapidly the study of interstellar molecules is moving away from simple discoveries into applications which can reveal otherwise hidden features of the way our galaxy is con-

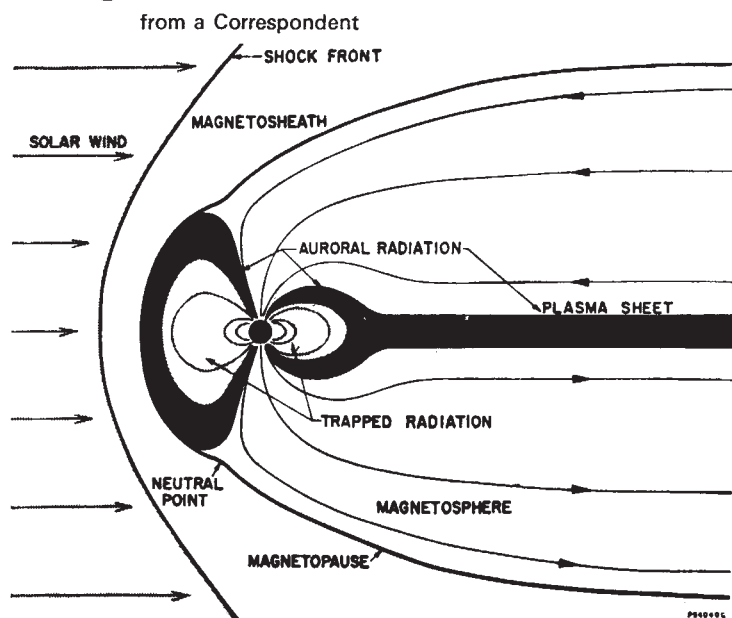
structed. One of the two sources definitely containing H^{13}CN is Sgr A, restoring the balance with Sgr B2 to some extent, and the other is Orion A. The two sets of observations give, by comparison with H^{12}CN emission from the same sources, carbon isotope ratios ($^{12}\text{C}/^{13}\text{C}$) of 4.7 and 8.3 respectively (*Astrophys. J.*, 163, L47; 1971). On Earth, this ratio is 89 and although, by themselves, these two measurements cannot be taken as meaningful for the galaxy in general, continued work along these lines will clearly help studies of our galaxy.

More work carried out with an NRAO 36 foot antenna, this time at Kitt Peak, by P. Solomon, K. B. Jefferts, A. A. Penzias and R. W. Wilson (the last two well known for their detection of the "3 K" background radiation) has also concentrated on using microwave molecular lines in measuring characteristics of a galactic source rather than looking for new lines. This study of the CO lines in the source IRC+10216 was inspired by the importance of this object at infrared wavelengths—at 5μ it is the brightest source outside the solar system, and has the large apparent diameter of $4''$ in the visible spectrum and $0.7''$ in the infrared. It is clear from the broad, flat nature of the CO line that it originates in a shell expanding away from a central object at 15 km s^{-1} . Extrapolation backwards in time suggests that this expansion started from the centre some 500 years ago, assuming that the shell was created in a single explosive event at the central, old carbon star associated with the nebula. But perhaps a more realistic process for the origin of this expanding source lies in the suggestion of Hoyle and Wickramasinghe that bright carbon stars must be continually losing mass as radiation pressure blows graphite outwards in a spectacular stellar wind (*Mon. Not. Roy. Astr. Soc.*, 124, 417; 1962).

So the study of interstellar molecules is rapidly forging a useful tool for the investigation of our galaxy. Isotopic abundances look likely to cause some puzzles, if they follow the lines of recent observations, and the evidence so far available does suggest that these abundances differ significantly from those in the solar system. A strange feature of the abundances of the molecules themselves lies in the way certain molecules seem to be favoured over others in the production mechanisms operating in the interstellar medium. Organic molecules seem to be favoured over simpler compounds such as NH_3 and OH, and the reason for this is at present a mystery. Making maps of the distribution of clouds containing complex molecules, in the same way that maps of hydrogen clouds have been made, will certainly improve our understanding of the geography of our galaxy. But in the public eye (and, indeed, to many specialists) this prosaic work is overshadowed by the growing conviction that clouds exist which contain even more complex molecules, including some, such as amino-acids, necessary for the development of life as we know it, and that these might be discovered at any time.

MAGNETOSPHERE

Convecting Field Lines



A model magnetosphere (meridional plane), according to B. J. O'Brien, showing the termination of the Earth's field at about ten Earth's radii towards the Sun.

A RECENT discussion meeting on the polar ionosphere and the magnetosphere, organized by the Royal Society on December 15 and 16, 1970, provided a forum for reviewing contemporary developments, both as regards magnetospheric models and dynamic processes, particularly those associated with substorms and their manifestations in the polar ionosphere. These latter seem to be caused by magnetic field lines convecting from the day to the midnight hemisphere where they re-connect and cause precipitation of energetic particles into the midnight auroral ionosphere. Although great magnetic storms are known to be initiated by increases in the solar wind flux and velocity, substorms occur in the auroral zone at other times. Thus of particular interest was the report by Dr P. J. Coleman (University of California, Los Angeles) of observations made on the satellite OGO 5 when, during a period of little or no change in the solar wind, the magnetopause boundary was observed to move in by two Earth radii. This accompanied a change in direction of the interplanetary field from northward to southward which was interpreted as causing connexion between the terrestrial and interplanetary field with subsequent convection of the field lines and erosion of the magnetopause boundary. Such a condition would facilitate entry of solar electrons and protons into the magnetosphere. Dr J. J. Quenby (Imperial College, London), on the other hand, reported calculations of particle trajectories which favour a sharp discontinuity between the magnetospheric tail and the interplanetary magnetic field.

The problem of the nature and role of

electric fields recurred throughout the meeting, particularly the question of whether there could be a significant component parallel to the magnetic field lines. Dr Pamela Rothwell (University of Southampton) pointed out also that abrupt changes in fluxes and pitch angle

distributions of gyrating energetic particles support the idea of longitudinal electric fields. Professor J. W. Dungey (Imperial College, London) felt that the conductivity along field lines was too high to support such fields, but he suggested that electromagnetic wave turbulence might provide a sufficient scattering mechanism to inhibit conduction.

Dr J. O. Thomas (Radio and Space Research Station, Slough) described the complex nature of the polar ionosphere and drew attention to the trough at about 60° magnetic latitude which seems to correspond with the plasmopause (the outer boundary of the relatively dense co-rotating magnetospheric plasma). Polewards of this, the Earth rotates under an ionospheric pattern including a relatively dense belt of ionization embracing the auroral oval and which is likely to be associated with enhanced fluxes of energetic electrons. A novel mechanism for the transfer of matter from the polar ionosphere into the tail of the magnetosphere was proposed by Dr T. E. Holzer (Imperial College, London). Neutral hydrogen escapes from the Earth's gravitational field at altitudes above about 500 km but the ionized constituents can escape only along the field lines. Holzer suggests that the space-charge electric field resulting from the separation of electrons and oxygen ions can accelerate hydrogen ions outwards to supersonic velocities.

A Million Years in Space

THE fall of a meteorite is still an important event for astronomers, as it always will be for laymen, but it is often months or years after a fall before all the information is gleaned from the fragments that reach the Earth's surface without having being burnt up in the atmosphere. An account of an analysis of a fragment of the Bovedy meteoroid which broke up in the atmosphere and scattered pieces over Northern Ireland (and possibly into the surrounding Atlantic Ocean) in the evening of April 25, 1969, is included in next Monday's *Nature Physical Science*. The report describes how the low level of gamma ray activity from a handful of isotopes in the fragment was measured at the Centre des Faibles Radioactivités of CNRS. The unexpected result reported by the group, J. Tobailem, D. Nordemann and T. Grjebine, is that the activity of ^{26}Al is surprisingly low, and the question is what this implies about the history of the meteoroid.

In meteoroids such as Bovedy this isotope of aluminium is produced chiefly by the action of cosmic rays on aluminium and silicon, so it ought to be possible to estimate the length of time for which the meteoroid was exposed to cosmic rays in space after its formation and before it

struck the Earth, assuming, of course, that the low level of activity does not mean simply that the fragment examined at CNRS came from within the meteoroid and was therefore shielded to some extent from cosmic rays. There are ways of allowing for the effects of shielding, however, and, using these, Tobailem *et al.* put forward an exposure age of 0.9 ± 0.3 million years. Presumably this corresponds to the time which elapsed since the Bovedy meteoroid broke away from the parent body. This value of exposure age given by the CNRS group does not seriously contradict an earlier measurement of the length of time during which the Bovedy meteoroid was exposed to cosmic rays, based on an examination of the rare gas content, but the earlier measurement also allowed an exposure age as long as 20 million years. The shorter ages allowed by the rare gas measurements were then thought to be spurious. It now seems that there is more to be said for the shorter ages, but these are considerably less than the ten to fifty million years which are the exposure ages of most stony meteorites. As Tobailem *et al.* say, this suggests a rather complicated history for the Bovedy meteoroid.

Several speakers dealt with the role of wave phenomena, their generation and their relation to particle precipitation. Both micropulsations (fluctuations in magnetic field strength in the frequency range 0.1 to 1 Hz) and very low frequency electromagnetic waves in mid-latitude seem to be equatorially generated and observations generally seem to support wave growth associated with anisotropy in the particle pitch angle distribution as suggested by Kennel and Petschek. High latitude v.l.f. emissions, however, occur on open field lines in longitudinal zones which seem to be related to the plasma sheet in the magnetospheric tail and to the neutral points on the solar side. A possible mechanism may be Čerenkov radiation from particles entering the polar ionosphere with velocity greater than the local phase velocity of the electromagnetic waves.

CHANDLER WOBBLE

Earthquake Correlations

from our Geomagnetism Correspondent

THE origin of the Chandler wobble, the precession of the Earth's axis of figure about its axis of rotation, has taxed the ingenuity of scientists ever since the phenomenon was discovered in 1891. But very little real progress has been made during the intervening eighty years. The most recent suggestion is that the random oscillations are maintained by the energy from earthquakes. Mansinha and Smylie (*J. Geophys. Res.*, **72**, 4731; 1967), for example, showed that very large earthquakes (magnitude greater than 7.5) correlate quite well with sudden changes in the centre of wobble during the period 1957–1967; and they took this to be an indication of a physical connexion between Chandler wobble and earthquakes. Hopes of this faded somewhat, however, when Ben-Menahem and Israel (*Geophys. J.*, **19**, 367; 1970) claimed that although, under favourable conditions, a single earthquake of magnitude 8.5 could maintain the Chandler wobble for about a year, all the real earthquakes taken together could only account for about 30 per cent of the observed wobble amplitude.

New evidence from Myerson (*J. Geophys. Res.*, **75**, 6612; 1970) has now modified the area of disagreement considerably by reaffirming the wobble-earthquake correlation (albeit in a different form), but at the same time suggesting that there may not, after all, be a direct causal connexion between the two phenomena. What Myerson has attempted is not a correlation between specific polar breaks and specific earthquakes, but a more general correlation between yearly means of the Chandler amplitude and yearly earthquake counts since about the beginning of the century. His technique

is, of course, statistical; but—to cut a long development short—Myerson treats the build-up of Chandler wobble as a random walk problem on the assumption that the excitation is by sudden, random breaks in the centre of wobble. Such breaks could, of course, be caused by earthquakes; but there is nothing in Myerson's development which presupposes this to be so.

What emerges at the other end is a good correlation between a function of the Chandler amplitude (actually the rate of change in the Chandler amplitude squared) and the yearly count of earthquakes with magnitudes greater than 7.0. There is thus no doubt that earthquakes are associated with wobble excitation. Indeed, this demonstration is more convincing than Mansinha and Smylie's simply because it is more general. There is, on the other hand, a very poor correlation between the same function of Chandler amplitude and yearly counts of earthquakes with Richter magnitudes greater than 7.5. Clearly therefore the relationship between earthquakes and wobble cannot be a simple cause and effect.

But finally, and perhaps most significantly, Myerson demonstrates a good correlation between the function of Chandler amplitude and the yearly count of deep and intermediate earthquakes

(magnitude greater than 7.0, depth greater than 70 km). This is likely to be a clue to the whole affair. It suggests that there may well be a deeper mechanism which both triggers earthquakes and maintains the Chandler wobble.

TRIBOLOGY

Interacting Surfaces

from a Correspondent

TRIBOLOGY, the study of the interactions between surfaces in relative motion, has become an international subject of great interest to chemists, physicists, and metallurgists, as well as to the mechanical engineers who started it all. This was evident from the wide-ranging interests and nationalities of more than a hundred participants at a conference on the chemistry of bearing surfaces held in the Swansea Tribology Centre on January 5 and 6.

In his introductory address, Dr D. Tabor (University of Cambridge), reviewing the present "state of the art" in tribology, pointed out that wear, which is more important than friction, can only take place when actual contact occurs between the sliding surfaces. The role of a lubricant, whether it be a gas or a liquid, is to prevent this contact. What happens when contact does occur? This

Do Earthquakes depend on Rock Type?

FROM an analysis of the worldwide distribution of earthquakes, Scholz (*Nature*, **221**, 165; 1969) concluded recently that although high differential motion is a necessary condition for the occurrence of earthquakes, it is not sufficient in itself. His argument was that there are many places in the world where the deviatoric stresses directly responsible for earthquakes are present but where no seismic activity results. In California, for example, no earthquakes have been observed below about 20 km and yet the necessary stresses must exist below this depth. Indeed, almost all seismic energy is released in the crust. To be sure, intermediate and deep earthquakes do occur; but they are almost always concentrated in narrow planar zones which dip at angles of about 45° below island arcs. It is clear that much of the mantle flows aseismically.

What, then, is the basic difference between the seismic and aseismic stressed regions which makes them one or the other? According to Scholz, seismic activity only occurs generally where the rocks are relatively acidic—a suggestion which seems to agree pretty well with both terrestrial and laboratory data. It has been found experimentally, for example, that the ease with which plastic

flow occurs increases with decreasing silica content—that is, with decreasing acidity. And it is a fact that the primary difference between crustal and mantle materials is the smaller proportion of silica in the latter. Scholz's hypothesis thus seems to be in general accord with what is known of the Earth's composition.

In next Monday's *Nature Physical Science*, T. Hatherton offers more specific evidence for the validity of Scholz's idea with data from New Zealand and the adjoining Tonga and Kermadec island arcs. It is a particularly convenient region to choose because of the contrasts involved. In the Tonga and Kermadec areas the axis of the arc neatly separates the oceanic crust to the east and the continental-type crust to the west. In accordance with Scholz's hypothesis it also neatly separates the seismic from the aseismic region. And if this were not evidence enough, in the New Zealand area where continental crust occurs on each side of the arc axis, so do the shallow earthquakes. The point is, of course, that plate tectonic theory implies that the deviatoric stresses in the lithosphere are symmetrical about island arc axes. Hatherton demonstrates that this symmetry only applies to earthquakes when all the rocks concerned are acidic.

was the question which most concerned the participants at the conference.

The chemists seemed to be interested chiefly in the effects of extreme-pressure additives on the load-carrying capacities of oils. For instance, Dr E. S. Forbes (BP Research Centre, Sunbury-on-Thames) demonstrated that the load-carrying ability of the alkylammonium dialkyl phosphates and dithiophosphates depended markedly on their structure. Dr E. V. Zaretsky (NASA Lewis Research Centre, Cleveland) described a fourteen-fold increase in life obtained by adding a substituted organic phosphate to a actual ball bearing being run with synthesized hydrocarbon oil under a low oxygen environment and at extreme pressures. This was a vivid illustration of the improvement obtainable by forming chemically a protective film between surfaces loaded beyond that point at which normal lubrication breaks down.

The presence of oxide films on the surfaces of worn metals was the subject of several contributions. Oxide films clearly have a protective role in that they prevent intermetallic contact. The oxidation of the metal, however, must also lead to an increased wear, if the oxide film cannot remain intact during sliding. Drs T. F. J. Quinn (University of Aston in Birmingham) and N. Tenwick (Queen Mary College, London) dealt with particular oxidation wear models. Both these workers underlined the need for a thorough examination of the kinetics of oxidation during wear, because the results of static oxidation experiments are apparently not relevant to the conditions occurring at the wearing interface.

The extremely reactive nature of clean metal surfaces (such as one has when the protective oxide film breaks up under cyclic mechanical or thermal stresses caused by sliding) was emphasized by Dr D. W. Morecroft (Shell Research, Thornton, Chester). He described some high vacuum experiments in which typical hydrocarbons were shown to react much more quickly with an oxide-free surface of iron than they would have done on the normal oxide-covered surface.

PALAEONTOLOGY

New Techniques

from a Correspondent

THE annual Christmas meeting of the Palaeontological Association held in Belfast from January 5-7 firmly established that the renewed vitality of the palaeontological sciences owed much to the application of new techniques from peripheral disciplines. The small size of the meeting alone acted as an effective means of furthering communication between active workers involved in interdisciplinary approaches from Britain, Ireland, Europe and North America.

The sixteen contributions covered a wide range of topics outlining the consequences of, and future developments arising from, isotope studies, electron microscopy, organic geochemistry, biochemistry, biological cultures, films of behaviour and numerical studies. The programme effectively divided into two spheres of interest: the functional and environmental, and the numerical. The key trend in developmental studies arising from all these approaches is towards a detailed understanding of the environments in which the fossils lived, secreted their skeletons and were buried, and the subsequent changes which affected them and their matrices. By contrast the chief advances in numerical studies are largely directed towards data storage, retrieval and evolutionary prediction.

Two encouraging trends emerged from a lively discussion from the floor. First, after years of separatist specialization,

palaeontologists are finding that trends in skeletal development, as revealed by combined mineralogical, biochemical and physiological approaches, are yielding a uniform end-product irrespective of phylum. Thus a common language and understanding of processes is developing as was shown by a comparison of investigations on the "shell" structures of brachiopods (Professor A. Williams, Queen's University, Belfast), bryozoa (Dr R. Tavener-Smith, Queen's University, Belfast) and molluscs (Dr J. Kennedy, Oxford). These studies use a combination of ultrastructural and light microscopic techniques in the comparison of the role of Recent soft tissues in calcification and diagenesis with well preserved fossil material which elucidate phyletic trends and classificatory problems.

Second, after several years of quantitative bewilderment, it seems that the application of computers to palaeontolo-

Fluctuations in Seismic Activity

IN next Monday's *Nature Physical Science*, G. F. Davies and J. N. Brune return to a problem which first attracted the famous seismologist Hogo Benioff just twenty years ago. Using an improved technique, they have apparently succeeded in detecting significant fluctuations in seismic activity over the seventy years or so since accurate measurements of earthquake magnitude were begun.

Benioff's original analysis involved calculation of the cumulative strain released by the world's large earthquakes between 1904 and 1951 from the sum of their energies. The result showed that during the 46 year period there had been five active phases separated by quiescent periods and that the cycles thus produced had become successively shorter. This was, and remains, a surprising conclusion. The deep seated global processes which account for seismic activity presumably have characteristic time scales running into millions of years. How, then, could they exhibit periodicities of a few tens of years or even less?

It was partly because of the unsatisfactory nature of Benioff's conclusion, and subsequent criticism of the way it was arrived at, that Davies and Brune have carried out a new analysis using a different technique. Their method involves the calculation of slip rates in fault zones by summing the seismic moments of the large (Richter magnitude greater than 7.0), shallow (depth less than 60 km) earthquakes within them. In essence, this is very little different from the Benioff method. Where it scores, however, is that the relationship between seismic moment and magnitude is much simpler than that between seismic energy and magnitude, and so fewer assumptions are

required. Thus, for example, whereas Benioff was strongly criticized for assuming that earthquake volume is independent of magnitude, Davies and Brune have neatly sidestepped the problem by effectively eliminating it.

Not surprisingly, the new analysis fails to confirm the existence of five cycles. What it does indicate, though, is a five-fold decrease in seismic activity since 1897. Indeed, the global data suggest three distinct periods of activity marked by well defined discontinuities. So although there seem to be no periodicities as such, Davies and Brune, like Benioff, are also faced with the problem of reconciling vastly different time scales, once accepting that the decrease with time is statistically significant. This they do by invoking mechanical coupling through lithospheric plates—an option which was not, of course, open to Benioff. In other words, they seek to explain the fluctuations in seismic activity by suggesting that the largest earthquakes are not independent.

The second result obtained by Davies and Brune clearly gives them greater satisfaction. Most of the slip rates they have calculated for particular seismic regions are agreed in general with the rates calculated independently by Le Pichon (*J. Geophys. Res.*, **73**, 3661; 1968) from plate tectonic theory. Obviously this is very satisfactory indeed, though in view of the many simplifying assumptions Davies and Brune still found necessary, it is also quite surprising. Even more surprising is the fact that the slip rate for the past twenty years is almost exactly equal to the average rate over the past ten million years obtained by Oliver and Isaaks (*Canad. J. Earth Sci.*, **5**, 985; 1968).

gical problems may be nearing a practicable solution (Dr P. H. A. Sneath, University of Leicester). So far stress has been laid on digital computation and the problem of defining suitable parameters in fossil materials (Dr J. S. Hampton, University of Lancaster), but Dr D. Raup's (University of Rochester) eloquent cinefilm of the analogue computation of simulated shell form seemed to present, however, a previously overlooked and crucial intermediary step between classical taxonomy and digital programming as exemplified by Dr J. L. Cuthill (Cambridge). Hence the ASCOP computer facilities established in 1961 by the Institute of Geological Sciences may well find themselves in demand by palaeontologists in the future.

Dr G. E. Farrow (University of Hull) showed that comparative studies of the Recent bivalve *Ceratoderma* (*Cardium*) *edule* and ancient analogues are beginning to rationalize some long standing palaeo-environmental arguments on the meaning of variations in shell sizes and structures. This work illustrates the biological deficiencies of many previous palaeontological reconstructions. Once again technological and molecular advances have overtaken biology before the palaeontologists' needs were fulfilled.

Isotopic studies were reviewed by Dr J. D. Hudson (University of Leicester) and exemplified in the Post-Pleistocene peat studies by Dr A. G. Smith (Queen's University, Belfast) and the Wealden palaeoenvironmental analyses of Professor P. Allen (University of Reading). Though promising, this field is not without difficulties, particularly economic ones.

In summation, although technological advances have made a significant contribution to palaeontological studies, they have, until recently, so outstripped palaeontological thought that many confusing and ill conceived papers resulted. These brought ridicule to the implementation of the techniques and thereby hampered palaeontological advances. This meeting showed, however, that a new era of thoughtful experimentation and expression accompanied by a renewed vigour is firmly established.

SURFACE CHEMISTRY

Sorption Phenomena

from a Correspondent

THE interdisciplinary nature of the study of gas/solid interfaces was clear when members of the Vacuum, Thin Films and Surfaces Groups of the Institute of Physics and the Physical Society met at the University of Liverpool on January 6. For many years, surface chemists have interpreted a wide variety of experimental data on chemisorption using a picture which describes chemisorption as the forma-

tion of a local molecular structure, involving the adsorbate, and a few substrate atoms, very similar in essence to an ordinary molecule. Dr T. B. Grimley (University of Liverpool) explained how this picture of a surface molecule is contained in a general quantum theory of chemisorption, and stressed the importance of experimental methods designed to investigate the energy levels of electrons in such surface molecules. To illustrate the theory, Grimley presented numerical results for sodium, barium and sulphur atoms on the (100) faces of tungsten and nickel.

One experimental technique for investigating the energy levels of electrons in ordinary molecules is photoelectron spectroscopy. A monoenergetic photon source (the helium resonance line, or the Mg K α X-radiation for example) ejects an electron from the target molecule with kinetic energy equal to the difference between the photon source energy, and the binding energy of the electron in the molecule, so that a measurement of the kinetic energy of the ejected electron yields the binding energy. Some of the results obtained were reviewed by Dr M. Barber (AEI Scientific Apparatus Ltd, Urmston), but the application of this technique to the study of adsorbed species is only just beginning. Its importance is that it

gives direct information on key theoretical quantities.

Two other experimental techniques were discussed. Professor J. H. Leck (University of Liverpool) presented some results obtained with electron bombardment desorption (EBD), and Dr J. C. Riviere (AERE, Harwell) described some recent developments in Auger spectroscopy. In EBD, a monoenergetic electron beam (15–200 volts) ejects electrons from adsorbed molecules, and positive ions, for example O⁺ from adsorbed carbon monoxide, which desorb, can be studied. The situation is qualitatively similar to electron-molecule collisions in the gas phase, and the method yields information on the structure and bonding of surface molecules. Tightly bound species are, however, inactive in EBD; in the tungsten/oxygen system for example, O⁺ is not desorbed at low oxygen coverages. No such limitation exists with Auger spectroscopy, but here Riviere reported the unwelcome observation that the primary electron beam can interfere with the adsorption process; it causes enhanced adsorption of CO on silicon substrates. This is a reminder that sorption phenomena are likely to be just as complicated in 1971 as ever before, and the point was emphasized when Mr R. G. Forbes (University of Cambridge) ex-

Drugs and Brain Tryptophan

MANY drugs are known to increase the rate of synthesis of the neurotransmitter serotonin in the brain. These include (+)-amphetamine, reserpine, lithium salts in large quantities and intraventricular injections of dibutyryl cyclic AMP; serotonin synthesis is also accelerated in the brains of animals exposed to high environmental temperatures. Conversely, the administration of para-chlorophenylalanine inhibits the synthesis of brain serotonin. It has generally been assumed that such effects are mediated directly or indirectly through some influence on the enzyme tryptophan hydroxylase which catalyses the rate-limiting step in serotonin biosynthesis, the conversion of tryptophan to 5-hydroxytryptophan. The report by Tagliamonte *et al.* in next Wednesday's *Nature New Biology*, however, suggests an alternative and novel mechanism by which drugs may influence serotonin synthesis. These authors report that the administration of (+)-amphetamine, reserpine, lithium salts or dibutyryl cyclic AMP all caused significant increases in the concentration of tryptophan in rat brain. Exposure of animals to high environmental temperature (40° C) also led to a large increase in brain tryptophan concentration. These increases ranged from a 60 per cent rise with reserpine to 300 per cent in rats exposed to 40° C.

The increases in brain tryptophan did not simply reflect changes in the concentration of the amino-acid in plasma, because reserpine and dibutyryl cyclic AMP caused increases in brain tryptophan without any significant increase in plasma tryptophan. Exposure of animals to cold stress, or the administration of a variety of other psychotropic drugs, failed to influence either brain or plasma tryptophan concentrations. On the other hand, the administration of para-chlorophenylalanine led to decreased concentrations of brain and plasma tryptophan.

Because the concentrations of tryptophan in mammalian brain are below the Michaelis constant of tryptophan hydroxylase, the rate of serotonin synthesis in the brain may be very dependent on the availability of the amino-acid. It should be remembered, however, that the concentration of tryptophan in the whole brain may not necessarily reflect accurately the intracellular concentration of this amino-acid in serotonin containing neurones. Nevertheless, the results of Tagliamonte *et al.* suggest that changes in brain tryptophan may well be important factors in controlling the synthesis of serotonin. The question of how drugs and high environmental temperatures cause changes in brain tryptophan remains unsolved.

plained why, in the field-ion microscope, the image gas is now thought to be strongly bound to the metal tip. Two years ago E. W. Müller thought that field ionization occurred at a clean metal surface, but his development of the atom-probe field-ion microscope has changed all that.

PLANT PATHOLOGY

Systemic Fungicides

from a Correspondent

FOUR years ago there were no commercially available systemic fungicides, whereas today there are ten of these compounds in practical use. At a conference organized in London on January 5 and 6 by the Pesticides Group of the Society of Chemical Industry, there was general agreement among the participants that these compounds are here to stay for a long time to come.

Systemic fungicides are applied to the soil or are sprayed on the leaves to protect crop plants from fungal diseases. They are a great advance on the old established method of treating the plant after a disease has started to develop, but little, however, is so far known about their mode of action. In his discussion of the advantages to be obtained from the proper use of efficient systemic fungicides Dr E. Evans (Fison's Agrochemicals) pointed to the danger that disease organisms might develop resistance to these specialized compounds. This question of resistance arose many times during discussions and there is now no doubt that, though systemic fungicides are a welcome addition to the armoury against plant disease, they will have to be used with care if widespread resistance by pathogens to specific compounds is to be avoided.

Although systemic fungicides will probably find their greatest outlet in staple food production, they have a place in other branches of agriculture, and two contributions by Dr R. J. Byrde (Long Ashton Research Station) and Miss Pauline M. Smith (Glasshouse Crops Research Institute) dealt with the assessment of their value for use on top fruit and on glasshouse crops respectively. Dr Byrde showed that benomyl, the thiophanates and triarimol were usually better than the standard captan/binapacryl spray for the control of apple scab and mildew, though they did not seem to depend on systemicity for their effectiveness. The monoculture system in the protected environment of the glasshouse presents special problems which Dr Smith and Dr D. M. Spencer (Glasshouse Crops Research Institute) kept in mind when designing experiments to evaluate ten currently available compounds for use on tomato and cucumber crops.

On the second day Dr T. W. Tanton (University of Southampton) said that the systemic fungicides now being developed seem to follow the path of solutes and water in the transpiration stream. They are therefore translocated passively in the xylem and free space within the tissues of treated plants and he described an elegant technique using lead-EDTA as a tracer to study distribution within this system. When the lead chelate, which is relatively non-phytotoxic, is precipitated *in situ* with hydrogen sulphide, studies can be made of the route followed by a compound applied to the soil, from root hair to regions of water loss at the leaf surface.

Although it has long been expected that systemic fungicides applied to any part of the living plant would move freely to other parts of the plant and control disease whenever it appears, such compounds are not among the first generation systemic fungicides. They move only in the direction of the transpiration stream and are not satisfactorily systemic in woody plants. Ethirimol and dimethirimol are taken up and translocated more rapidly by cucumber than by apple as Dr Claire Shephard (Plant Protection Ltd) has found. Dr R. T. Mercer (May and Baker Ltd) dealt with three thiophanate compounds, one of which, thiophanate-methyl in water, gives rise to methyl benzimidazole-2-carbamate, a fungitoxic molecule which is also formed

from benomyl. The uptake and translocation in barley seedlings of 'Cela 524', a piperazine compound, have been studied by Dr V. von Bruckhausen (Boehringer Sohn) and his colleagues using inhibition of spore germination, the bioautography of thin layer chromatograms and the incorporation of the tritium labelled molecule into plant tissues as measures of uptake.

In a final discussion which ranged widely over such topics as the areas of need for systemic fungicides, their present usage and their mode of action, the phytoalexins, the naturally occurring antifungal agents, were mentioned as other possible systemic fungicidal molecules. There is an obvious need for much more research on the chemical basis of disease resistance.

CIRCADIAN RHYTHMS

Keeping in Time

from our
Experimental Psychology Correspondent

THOUGH light is the most important of the signals that synchronize circadian rhythms in animals, in man, under properly controlled conditions, the lighting cycle is relatively ineffectual. This conclusion (R. Wever, *Pflügers Arch.*, **321**, 133; 1970) is one of several interesting results on human diurnal rhythms

Revealing DNA in Chromatin

CHROMATIN—the complex of nucleic acid and protein which is obtained when DNA is extracted from the cells of higher organisms—is usually thought to have a structure in which the DNA is extensively, if not completely, covered by basic histone proteins. This idea is now shown to be incorrect by Clark and Felsenfeld who report in next week's *Nature New Biology* the application of three fairly straightforward analytical techniques to chromatin of calf thymus.

The saturation value reached when chromatin is titrated with polylysine indicates the amount of DNA which is free to react with the polypeptide. Measurement of the amount of DNA remaining after chromatin has been digested with staphylococcal nuclease shows how much DNA is protected from the enzyme. Both results suggest that roughly half of the DNA of chromatin is not protected by its associated proteins from reactions.

Another surprise has emerged from studies to find how tightly the proteins of chromatin are bound to DNA. The technique used by Clark and Felsenfeld was to mix chromatin with radioactively labelled free DNA and then to digest the mixture with nuclease. Any radioactivity which is not digested must have been

protected by proteins which were free to exchange between the free DNA and the DNA of chromatin.

The answer obtained depends on the conditions used to incubate the mixture of DNA and chromatin. At the ionic strengths used to measure the reaction of chromatin with polylysine or with nuclease, almost all the radioactivity is digested. But as the ionic strength is increased, more of the DNA is protected against digestion. This means that many of the chromatin proteins must in these conditions be in labile equilibrium with DNA, and not tightly attached as had been previously thought. Unfortunately, many experiments with chromatin have used these latter conditions, so that their conclusions must now be regarded as suspect.

When chromatin is transcribed by RNA polymerase, only 5–20 per cent of the DNA is transcribed into RNA, but much more of the DNA is transcribed when the histone proteins are removed. This has usually been interpreted to mean that histones function in some way as gene repressors which must be removed to enable genes to function. The implication of Clark and Felsenfeld's experiments is that, for much of the DNA at any rate, this type of model is probably irrelevant.

now coming from a group at the Max Planck Institute at Seewiesen and Erling-Andechs. Wever proposes that in man opportunities for social interaction are more important synchronizing agents (*zeitgebers*) than any purely physical stimuli.

In his experiments, human subjects, either alone or in pairs, lived for several weeks in an underground isolation chamber, and were subjected to a 12 h light : 12 h dark cycle of artificial illumination. This was the only source of external timing, and all subjects failed to become entrained by it. Their rhythm of sleep and wakefulness and their rectal temperature cycle lagged behind the lighting cycle, drifting away from it with a phase difference of about an hour a day. In contrast, subjects who in addition were summoned by the sound of a gong at regular intervals to give urine samples and to perform a series of tests, all became entrained, and indeed changed phase appropriately in response to 6 h phase shifts in the imposed 24 h rhythm.

Non-linear oscillators are believed to be responsible for circadian rhythms. The theory of such mechanisms makes it clear that even very weak coupling of an oscillator to a periodic source allows entrainment. Even so, light as such is evidently too weak to act as a *zeitgeber* in man. Though Wever's results do not indicate what the most effective periodic stimuli might be, it seems very likely that alarm clocks, going to work, lunch time, and the like, by virtue of their social and psychological significance have become the periodic events by which the biological clocks of man are set.

MULTIPLE SCLEROSIS

Myelin Milestones

from our Molecular Biology Correspondent
THE study of demyelinating diseases is an area in which protein chemistry is now skating very close to the surface of medicine. In the experimentally induced condition, allergic encephalitis, an animal after injection with a myelin constituent becomes sensitized against its own central nervous tissue. The fraction with this cataclysmic activity is the basic protein A1, which is one of the two major protein components of the membrane, of which it comprises some 30 per cent. It is also known that a nonapeptide fragment of this protein suffices for activity, and according to Westall *et al.* (*Nature*, **229**, 22; 1971) A1 from animals in which the crucial residues, tryptophan, glutamine and lysine or arginine, are absent from this part of the chain also possesses no encephalitis-inducing activity. Westall *et al.* have further demonstrated the absolute requirement for tryptophan, followed after four residues by glu.lys or glu.arg, by testing a series of twelve synthetic peptides.

The sequences of complete A1 chains have now been reported by Eylar (*Proc. US Nat. Acad. Sci.*, **67**, 1425; 1970), and by Carnegie (*Nature*, **229**, 25; 1971). Eylar has determined both the bovine and the human sequences, which have 170 and 172 residues, and differ from one another at twelve points, including a dipeptide insertion in the human protein. The vital tryptophan residue is at position 116 (referred to the bovine sequence). Carnegie's sequence of human A1 is not quite complete, and, moreover, differs from that of Eylar at several points, and grossly in one tryptic peptide. Both authors agree that a previously reported electrophoretic heterogeneity may arise in part from unusual modifications in one arginine residue.

It has also now emerged that the tryptophan-containing sequence may not be the only lethal site. In guinea-pigs, specific modification of the single tryptophan leads to inactivation. In rabbits, however, the remote peptic peptide, 43-91, causes encephalitis. This is confirmed by Chao and Einstein (*J. Biol. Chem.*, **245**, 6397; 1970), who find that all of several chemical modifications of the tryptophan lead to inactivation towards guinea-pigs, whereas activity towards rabbits is retained. They refer in

passing, moreover, to a striking finding, whereby guinea-pigs, injected with the modified, inactive material, become resistant to the effects of the native A1 protein.

One must suppose that A1 must play some essential structural part in the membrane. It differs from most proteins found in membranes, which tend to be predominantly acidic, and is apparently a random coil in the extracted state. Eylar notes that the basic amino-acids are spread out along the chain, and that there are five clusters of non-polar and negatively charged residues in the sequence, one of which contains the tryptophan site. So far there seem to be few ideas about the mechanism of the auto-immune process, though Carnegie offers a speculation, based, however, on unconvincing structural arguments, whereby 5-hydroxytryptamine interacts with the vital tryptophan and a nearby phenylalanine—though the latter, as Westall *et al.* show, is unnecessary for activity. The suggestion then is that this binding site is blocked by an immune interaction. The assumption in all this work is that the experimental condition is related in its causes, no less than in its manifestations, to the real human disease.

More about Mycoplasmas

OUTSIDE the world of veterinary medicine mycoplasmas are a comparatively ignored group of organisms. They have a certain curiosity value as the smallest free living organisms known, intermediate in size between viruses and bacteria, and they are often a cause of concern in laboratories working with cultivated eukaryotic cells, cultures of which mycoplasmas often contaminate, but beyond that they are generally forgotten. Attitudes may change, however, because at least one form of arthritis in swine results from mycoplasma infection and the suggestion that arthritis in man may have a similar cause cannot be airily dismissed. Investigations of the basic biology of mycoplasmas such as those reported in next Wednesday's *Nature New Biology* by Slater and Folsome and Gourlay and his colleagues may therefore prove to be of more than just academic interest.

Slater and Folsome, working with *Mycoplasma laidlawii* A, have adopted the approach used to demonstrate inducible enzymes in bacteria to show that maltose induces an enzyme involved in its metabolism, α -glucosidase, in *M. laidlawii*. When mycoplasmas grown on a medium containing glucose are transferred to a medium in which maltose is the principal energy source, there is a lag before the cells begin to multiply in the new medium and to synthesize proteins including the α -glucosidase. Once the lag is over, however, the amount of this

enzyme in the cells induced by maltose is some ten-fold greater than in control cultures maintained throughout in a glucose-containing medium. And by comparing the specific activities of the α -glucosidase and other enzymes in cells growing in either of the two media Slater and Folsome have ruled out any suggestion that the apparent induction results from some trivial artefact. Furthermore, they have shown that glucose does not suppress the induction of this enzyme by maltose and that it is possible to isolate partially constitutive mutants.

Clearly, the expression of the genome of mycoplasmas is regulated and there may well be close parallels between the molecular mechanisms which effect such regulation and those acting in bacteria. Mycoplasmas also share another important characteristic with bacteria and other cells; they are susceptible to infection by viruses. Last year Gourlay reported (*Nature*, **225**, 1165; 1970) the first discovery of a virus which infects mycoplasma. He and his colleagues have now further characterized this agent. The virus particles appear as short rigid rods, about 150Å by 900Å, with rounded ends, and because they are labelled when they are grown in a medium containing tritiated thymidine but not when they are grown in a medium containing tritiated uridine, Gourlay and his colleagues believe they must have a DNA rather than an RNA genome.

PEPTIDES

Closing the Ring

from our Cell Biology Correspondent

THE tyrocidines and gramicidins produced by various strains of *Bacillus brevis* are exceptionally interesting molecules; they are closely related cyclic decapeptides which contain the D form of phenylalanine and ornithine, an amino-acid, which does not, of course, occur in proteins; their synthesis does not involve any ribonucleic acids and they have antibiotic properties. Because the sequence of the ten amino-acids in these molecules is not determined by a messenger RNA, the enzymes responsible for their synthesis have, not surprisingly, attracted much attention. And as a result of recent work, notably in Lipmann's laboratory and by Stoll and his colleagues in Oslo, we now have a clear picture of how the gramicidins and tyrocidines are synthesized; there are striking differences as well as basic similarities.

It has been known for some time that two enzymes, I and II, are required for the synthesis of gramicidin S. Enzyme II activates and racemizes L-phenylalanine residues, and enzyme I, the larger of the two molecules, activates L-proline, L-valine, L-ornithine and L-leucine and catalyses the formation of the peptide bonds. Earlier studies had shown that the largest peptide precursor of gramicidin S is a pentapeptide and, because the complete molecule comprises two repeats of the sequence D-phenylalanine-L-proline-L-valine-L-ornithine-L-leucine, it had been suggested that a molecule of gramicidin S is formed by the head to tail condensation of two such pentapeptides, catalysed by enzyme I.

Stoll and his colleagues in Oslo, in a neat series of experiments, have now confirmed this idea and have shown that the condensation of the two pentapeptides occurs on a single enzyme I molecule (*FEBS Lett.*, 11, 348; 1970). They have exploited the finding that the gramicidin synthetase accepts azetidine-2-carboxylic acid in place of proline. Enzyme I molecules charged with pentapeptides containing either proline or the analogue were mixed and synthesis of gramicidin was allowed to go to completion. If two enzyme I molecules each carrying one pentapeptide cooperate to produce the decapeptide, 50 per cent of the gramicidin produced by the mixture should be hybrid with both proline and azetidine-2-carboxylic acid residues. By contrast, if each enzyme I molecule carries two pentapeptides, which are condensed intramolecularly, no hybrid gramicidins should be formed and this turned out to be the case. The final step in gramicidin biosynthesis seems therefore to occur on a single enzyme I molecule. Stoll and his colleagues suggest that one pentapeptide sits and

waits at a site on the enzyme and the second chain is synthesized attached by a thioester bond to a phosphopantetheine residue in the enzyme. The two pentapeptides once completed can then be brought close enough to allow the head to tail condensation.

Synthesis of the tyrocidines, according to Roskoski and his colleagues (*Biochemistry*, 9, 4839; 1970), involves three enzymes with molecular weights of 100,000, 230,000 and 460,000. The light and intermediate molecules respectively activate phenylalanine and proline and the heaviest molecule activates the L forms of leucine, ornithine, valine, tyrosine, glutamine and asparagine, and is the site of peptide bond formation. As with gramicidin synthesis activation of each amino-acid involves splitting a molecule of ATP and the growing peptide chain is attached to the enzyme by a thio ester bond. Unlike gramicidin synthesis, however, the synthesis of tyrocidine involves the step by step polymerization of all ten amino-acids. Roskoski *et al.* (*ibid.*, 4846) have been able to isolate

the complete series, ranging from dipeptide to decapeptide, of linear precursors of tyrocidine. And apparently once the linear decapeptide has been made, it is comparatively slowly cyclized by the formation of a peptide bond between the amino-group of the free amino-terminal residue, D-phenylalanine, and the carboxy group of the carboxy terminal leucine residue, which is both activated or held to the enzyme by the thio ester bond.

In retrospect this striking difference in the final stages of the biosynthesis of these two molecules is not all that surprising because the decapeptide sequence of tyrocidine is unique rather than a repeat of two pentapeptide sequences. More surprising is the finding reported by Roskoski *et al.* that the phenylalanine activating and racemizing enzymes in the two systems, which play a comparatively small part in the biosynthesis, are not interchangeable. The ability to determine the sequence of a peptide without the involvement of RNA clearly has its price.

Isoenzymes of Choline Acetylase

A FURTHER twist to an already complex story is added by the report by Fonnum in next Wednesday's *Nature New Biology* of the existence of multiple forms of the enzyme choline acetyl transferase (ChAc) in the brain of the rat.

Following the discovery by Whittaker and De Robertis of the existence of pinched off nerve terminals, or synaptosomes, in homogenates of mammalian brain, many studies have made use of centrifugation techniques to determine the subcellular distribution of neurotransmitters and related enzymes in neurones in the central nervous system. There has been general agreement that most of the acetylcholine in brain is localized in nerve terminals, and that a substantial portion of this acetylcholine is stored within synaptic vesicles which can be liberated from synaptosomes by mild hypo-osmotic shock (Whittaker *et al.*, *Biochem. J.*, 90, 293; 1964). On the other hand, there has been considerable disagreement about the subcellular localization of the acetylcholine synthesizing enzyme ChAc in mammalian brain. Although this enzyme was specifically localized in synaptosomes, most of the synaptosomal ChAc seemed to be associated with synaptic vesicles in rat and rabbit brain, but in guinea-pig and pigeon the enzyme seemed to be located in the cytoplasmic fraction (McCaman *et al.*, *J. Neurochem.*, 12, 927; 1965; Tucek, *J. Neurochem.*, 13, 1317; 1966).

In an important series of studies Fonnum has illustrated some of the pitfalls which may befall even the wary in the interpretation of subcellular distribution results. Fonnum showed

(*Biochem. J.*, 103, 262; 1967; *Biochem. J.*, 109, 389; 1968; *J. Neurochem.*, 17, 1095; 1970) that the ChAc which seemed to be associated with synaptic vesicles after osmotic shock treatment of homogenates of rat and rabbit brains could be easily solubilized by increasing the pH and ionic strength to values similar to those likely to prevail intracellularly. Species differences existed in the ease with which ChAc non-specifically adsorbed to vesicle or other lipid membranes in media of low ionic strength. Such differences were shown to correspond to species differences in the surface charge of the ChAc molecules, those from rat and rabbit being more strongly adsorbed by cation exchangers than the enzymes from guinea-pig or pigeon.

In *Nature New Biology*, Fonnum shows that several forms of ChAc with different isoelectric points can be separated by isoelectric focusing of partially purified preparation of ChAc from rat brain. The three different forms of the enzyme, with isoelectric points in the range pH 7.5 to 8.3, were also demonstrated in crude extracts of rat brain.

ChAc from guinea-pig or pigeon brain, however, gave only single peaks of enzyme activity after isoelectric focusing, with isoelectric points of 6.7 and 6.6, consistent with the species differences in surface charge previously described. Further studies are in progress to determine whether the different forms of the enzyme in rat brain have the same cellular and subcellular distribution or whether they are present in morphologically or functionally distinct compartments.

Planning Complex Building Systems

by
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A criticism of current methods reveals the problems of planning a large building complex. The results of recent research on health sciences centres suggest that there may be a more profitable approach.

CONSIDERABLE emphasis is currently placed on traffic and communication analysis as a basis for the planning of large, institutional building complexes—notably hospitals and universities¹⁻⁶. Although such methods differ, they invariably conform essentially to the procedure outlined by Whitehead, Willoughby, Paterson and Drummond⁷. A matrix is arranged of the suitably identifiable activity units constituting the buildings being designed. A hospital operating theatre, for example, might typically be divided into some twenty such units. The matrix entries represent a measure of association between these units based on such data as the number of journeys made between them, the transportation of goods and so on. In short, some measure of the total information flows between departments. A computer then uses a heuristic procedure to group highly interdependent activity units and, on this basis, an approximate floor plan is developed*.

Early attempts at these methods were concerned with the relatively simple case of single storey layouts, but recent efforts are more realistic in this respect⁹. They can also contend with various site constraints. It can be specified, for example, which spaces are to be located on the site periphery for some reason. The problem is to collate all the designated spaces to achieve adjacent positioning where this is most highly rated from the activity matrix, that is, to maximize the sum of arc values of the graph representing this situation where the arc values correspond to the matrix weighting (Fig. 1). A sequence of the essential steps involved in the whole procedure is shown in Fig. 2.

I shall not enumerate the various methodological difficulties associated with these techniques; most are well documented in the literature of the subject as future problems to be solved. My purpose here is to make a more fundamental criticism. I have based this on consideration of a particular type of building—health sciences centres—but it applies generally to the design of buildings for complex organizations.

Health Sciences Centres

Health sciences organizations usually consist of a university teaching hospital, an associated medical school and several paramedical schools for education in the applied health professions. The principal functions of such organizations are:

* It is unfortunate that articles on traffic flow methods often give a quite false impression of the overall problems of physical planning. They are concerned solely with location rather than what to locate, in terms of both quantity and type of space. In the health fields, for example, it is frequently the case that far from providing the designers with a convenient list of departments, the responsible authority has only the vaguest idea on what facilities the building will eventually provide (ref. 8).

the education of students; the provision of health to the surrounding community; and medical research. They frequently maintain about a hundred departments and a resident population of three or four thousand.

Beer¹⁰ and others have shown how a large industrial company can usefully be considered as a homeostat and this terminology is easily extended to the health sciences situation.

The state of the health sciences system at any time can be measured in terms of such things as the number of available staff, the number of patients, the number of available rooms, and so on. Its output measure is the set of conditions which constitutes its "goals"—not to exceed its budget, to educate a certain number of students, and so on. Obviously all these aims cannot be optimized simultaneously. We can only see the "overall aim" of the system as that of being environmentally viable—to move towards stability in a homeostatic relationship with the environment.

The state of the environment of the health sciences system can be measured in terms of the requirements for trained doctors, the number of sick people, economic and resource availabilities, and so on. With the health sciences system and its environment homeostatically related, the state of the former's aims is the input to the environment while the environment's own states are the health sciences system's inputs.

What this means essentially is that a viable organization is sensitive to environmental changes and, in response to these as inputs, is continually trying to evolve into a different organization. Although the overall function of the organization may stay relatively constant, the ways in which it fulfils this function may not. This is particularly true in a thriving educational institution where, prompted by research results, new staff appointment and policies, uncertainty over grants and so on, major organizational changes can, and should, occur frequently.

We can imagine the state of the whole system described by the states $S_1, S_2, \dots, S_n$ of its S_n components. These values will determine a unique point in the n dimensional phase-space S such that there is an isomorphic correspondence between points in the space and the states of the system, the set of all

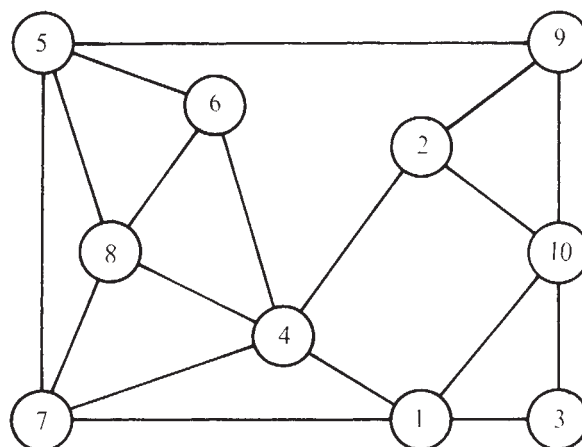


Fig. 1 The nodes of the graph correspond to the various activity units and the arcs to the degrees of association of such units from the matrix.

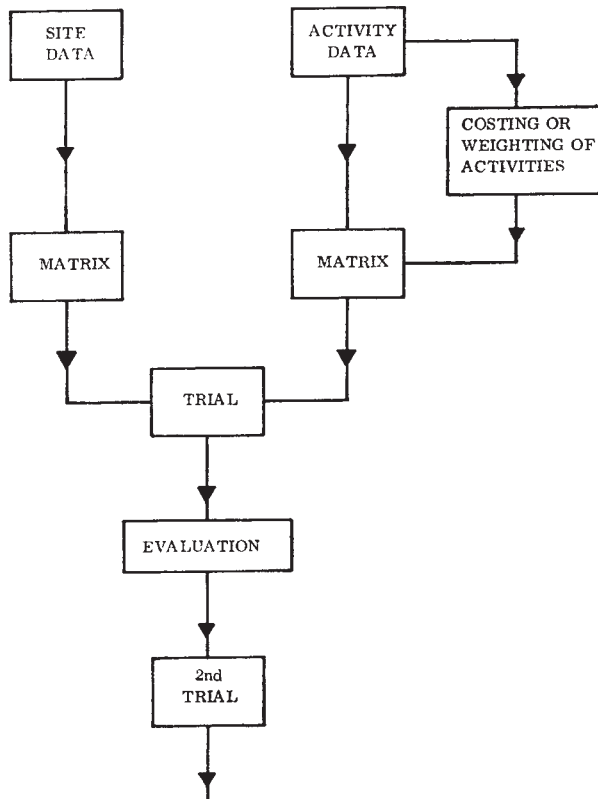


Fig. 2 Flow diagram of the essential stages involved in a typical programme to derive a building layout.

such points corresponding to the total possible states of the system.

As the component states $S_1, S_2, S_3, \dots$ change in response to inputs from the environment, so the point representing the state of the whole system will trace a curve in the n dimensional space. At discrete time intervals throughout the life of the system the location of this point will vary.

The objection to traffic and communication analysis can be clarified through such a description. What this procedure does is to speculate on the location of this point in the organizational phase-space at one point in time. The state of the system this represents is measured in terms of communications and the results are translated into the physical hardware of the building. Meanwhile, in the intervening years between the traffic study and the completion of the building, things have changed. The same funds are not available; a large research project in microbiology, say, has been cut back but money has become available from other sources for a specific programme in nuclear physics, not envisaged a year or two ago. The point representing the state the system now needs to assume has moved on, but the buildings which must accommodate this system have been designed around an earlier analysis. The state of the system is now artificially constrained to a region of the space although the maintenance of successful behaviour requires it to be somewhere else. The right facilities for current requirements are just not available and the building may well be inadequate before it is operational. The more detailed and "accurate" the traffic flow data and the more care taken in realizing the analysis in the building hardware, the higher is the probability of the building rapidly becoming obsolete.

These problems are by no means academic. The average duration of research projects, for example, is frequently less than four or five years but the life of medical school buildings is in excess of forty or fifty. In that time the buildings will be required to accommodate many different groups each with significantly different and varying requirements for space, servicing and communications with other groups.

Changes such as these depend to a large extent on the situa-

tion of a particular institution—the source of its grants, the prestige of its professors and their demands for space and equipment and so on—as well as the more general changes in educational and treatment policies operating on a national or international scale. During the past few years, for example, there has been almost universal acceptance of the central sterile supply department to replace individual sterilizing units at many points in the hospital. This has quite radically affected the planning of the hospital and a building planned rigidly to the old system has difficulty in adapting to such changes. At present almost all new hospitals are designed with central sterilizing departments and the planning layouts associated with them, but should regional centres eventually take over these functions, as is already happening in some places, then the central sterile supply department will disappear completely from the hospital and with it much of current hospital layout. Similar changes could occur if food were prepared outside the hospital or with the introduction of computer controlled patient monitoring systems.

In the medical school the breakdown in some places of the traditional divisions of teaching into preclinical, clinical and intern years and the emphasis on community-based medicine may have far-reaching effects.

Communication analysis is contributing little towards solving these problems—it does not prohibit the design of buildings which impose unnecessary constraints on their organizations. As I have shown, it even increases the chances of this occurring. In the terms used earlier, the operation of an organization within these constraints is homeostatic only in very restricted boundary conditions of input. Instead of an organization evolving to meet the requirements of its environment (that is, restoring stability) it changes in ways as much prompted and directed by what is possible for the buildings. The function of the designer should rather be seen as that of allowing extension of these boundary conditions in finding a suitable interpretation of "ultrastability"¹¹ in building terms.

This is not to say that traffic and communication studies are of no use. I think, however, that as presently carried out their emphasis is too deterministic to form the basis of a design methodology for systems which must contend with unpredictable future change.

Alternative Formulation of Design Problem

I have attempted to argue that the current emphasis on achieving optimal location in physical planning is mistaken. This emphasis ignores the dynamic character of the institutions which are being accommodated, and is more concerned with providing unique, best solutions to their requirements. Such an approach has some well established precedents in architecture. As self-adapting systems, such institutions and their requirements are constantly evolving. In these terms a

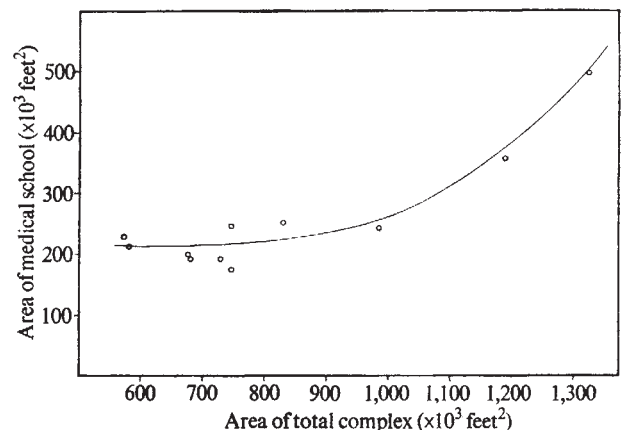


Fig. 3 The area of the medical school against the area of the total health sciences complex for 11 buildings at various stages of planning and development.

solution to the optimal layout problem is trivial. Its validity holds for only a small part of the time for which its physical realization in building form will operate.

The formulation of any general theory of the design of buildings for complex organizations must account for two requirements. First, there is the need for a method of planning which ensures that the building system incorporates properties valid throughout the transformations the organizational system will undergo in time. Second, there should be a physical interpretation of these properties in terms of building hardware which minimizes constraint on the organization being accommodated and allows it to locate and relocate its organizational subsystems with maximum flexibility.

The most notable attempts at satisfying the second requirement can be seen in the work of Weeks¹², who has argued that, although specific activities may have very specific environmental requirements, provision of these is not incompatible with a non-specific physical framework^{13,14}. In other words, it is possible to provide specialized activities with the facilities they require without committing all such activities to particular locations indefinitely.

The frequency distribution of room sizes in large institutional buildings¹⁵ has a sharp peak at about 150 feet²—a great many activities of the sort occurring in hospitals operate successfully in spaces of about this size. The variable factor in the provision of physical environment is not so much the room size but the requirements for electrical, piped and environmental servicing.

Weeks has taken advantage of this work; in the introduction to proposals for a new medical school he says

"A regular grid network of communications corridors will gradually be established as construction proceeds, which will serve buildings designed around a module of space which can be subdivided into a wide range of useful sizes of rooms and is serviceable in a very flexible way. In this way non-specific space is converted, by the installation of the services required, into specific space. This remains changeable, and occupancy of groups of spaces by a particular discipline or sub-discipline will not therefore inhibit change of function in the future."¹⁶

The hypothesis, then, is that the built-in potential for more mechanical and environmental servicing than is actually required at any one time allows the building to "respond" closely to changing organizational requirements. The cybernetic interpretation of this is, of course, that of incorporating sufficient redundancy to increase the systems ability to contend with the unpredictable.

Language for Planning

As complex organizations adapt to maintain goal achievement in a changing environment, their needs for accommodation will change. To delay obsolescence for as long as possible, buildings should have the capacity to "absorb" these changes.

To refer back to my first requirement of a general theory of design, the difficulty which faces the physical planner is that of

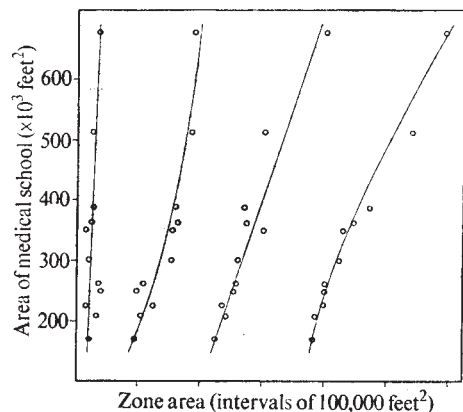


Fig. 4 The area of the four functional zones against the area of the medical school. The zones are uniformly spaced along axis for clarity on one graph. (Zones from left to right are: administration; shared facilities; clinical sciences; basic sciences.)

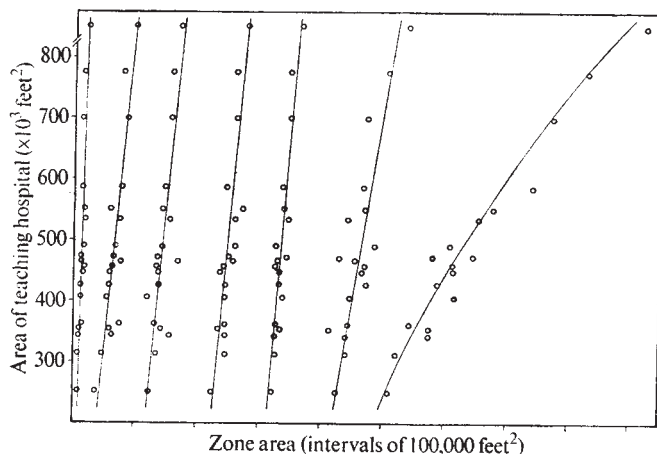


Fig. 5 The seven hospital zones against the hospital area. Again zones are spaced for clarity. (Zones from left to right are: administration; non-medical services; medical services; treatment; diagnostic; outpatients; inpatients.)

being able to plan facilities in the knowledge that the required specifications of these facilities will change, but without detailed knowledge of the form the changes will take. The state description of the building system will constantly change and yet we must have such a description in order to plan.

In an effort to look at the ways in which such a description might be expected to change, I have analysed data from a sample of eighteen university hospitals and eleven medical schools. The buildings were divided into eleven spatial components corresponding to the broad functional divisions within the organization. The medical school, for example, consists of four such zones—basic sciences, clinical sciences, shared facilities and administration. The seven hospital zones include those for treatment, diagnosis, back-up and support and so on. Each zone is further divided into departments and these in turn into sub-specialty groups and eventually into rooms which are classified again into one of three degrees of servicing. These range from the basic supply of electricity (office spaces and so on) to the supply of special gases and environmental control (laboratories and theatres). This hierarchical structure is proving useful in determining what the nature of a planning description could be.

Fig. 3 shows the relationship between the area of the overall health sciences complex and that of the medical school. The consistency reflects the many interdependencies. A medical school must be a minimum size to provide the necessary facilities for its particular number of medical students and the amount of research to be carried out. This number of students requires a minimum of beds of resident patients to provide a sufficient range of illness for their instruction so that, in turn, a specific number of operating theatres and a specific amount of back-up and servicing is required for a balanced hospital.

Fig. 4 shows the four functional zones of the medical school against the total medical school area. Again consistencies appear which reflect, for example, the balance of basic and clinical teaching and research providable under the current educational and economic constraints; the amount of administration necessary and so on. Fig. 5 shows a similar analysis of the hospital zones.

At a departmental level of analysis such relationships vary in consistency. Most hospital departments relate very well to their zone areas and to other departments. In the medical school, however, decisions of educational policy must be considered, in order to account for the variation in some departments. Nevertheless an analysis of even these departments based on amounts of space provided with certain degrees of servicing (Fig. 6) has considerable consistency. A biochemistry department in one health sciences complex looks very much like another when described by the relative balance of laboratory office and teaching space and so on.

This is the beginning, then, of the sort of language that could be used by the planner—description of a complex building as a hierarchic system allows considerable simplification. Equally important, however, is that such a description is applicable to many situations. All of the hospitals in the sample are different. They all operate in unique situations of locality and personnel. If, however, they are described as hierarchic systems, any of the hospitals in the sample could, by a suitable set of transformations applied to this description at the appropriate level of the hierarchy, be transformed into any other.

Recalling the phase space description which I used earlier, it would be best to be able to fix some of the coordinates to obtain a time-dependent measure of the system for which there existed measure-preserving transformation groups. Then transformation of the system S_1 (as defined by its group invariants) at time t_1 into itself at t_2 is equivalent to transforming the system into a second system S_2 at the same time t_1 .

Perhaps then it is a description of this nature which will best satisfy the requirements of a planning language, namely that it will apply throughout the detailed changes which a particular organization and its buildings will undergo.

Many analogies are applicable to this situation, for example members of a species. Their ability to deal with changing special or local conditions is a sort of "non-genetic" variation through which the individual adapts. This is analogous to the incorporation of redundancy in the "non-specific" physical framework of a building complex mentioned earlier. The planning situation, however, is more analogous to the "genetic" variation adapting the whole species to its larger environment¹⁷. Provision of an appropriate "genetic" structure by the planner allows a building to adapt at the individual level to many special requirements. I would not wish to take this analogy any further but it is interesting (Fig. 7) that the influences of two different national environments produce differences in some spatial relationships in health sciences systems fulfilling very similar functions.

These findings also fit in well with the conception of health sciences systems as homeostats. In systems which are more or less successful invariant relationships would be expected between some system variables and the systems goals, independent of environmental variations^{11,18,19}. In the case of a simple ultrastable device designed for a given purpose these variables would of course be known—that would be implicit in the specification. In a complex artificial system such as an administrative organization, however, these goals are largely undefined.

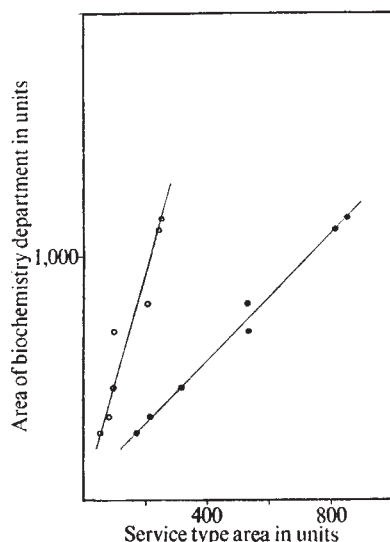


Fig. 6 A typical medical school department (biochemistry) showing amounts of floor area supplied with types 1 and 3 servicing. Type 1 (○) is principally office accommodation and type 3 (●) is laboratories. (Area is shown in units of 36 feet².)

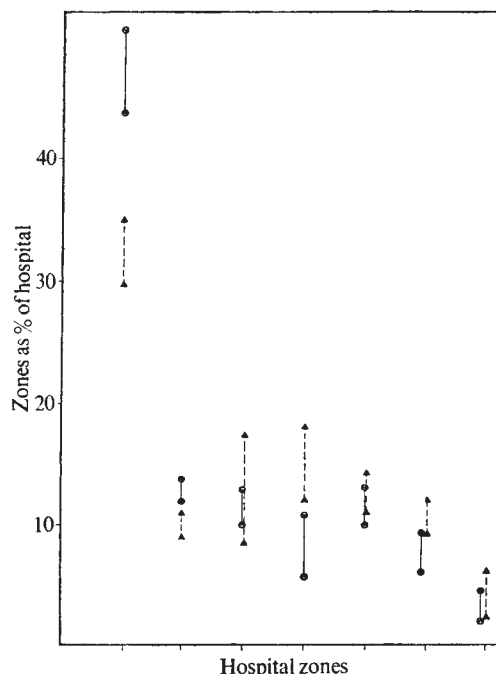


Fig. 7 The influences of two national environments on the relative balance of space provision. All North American health sciences centres fall within the broken line range shown (▲---▲); United Kingdom centres within the solid line (○—○). (Zones from left to right are: inpatients; outpatients; administration; non-medical services; treatment; diagnostic; medical support.)

Attempts to predict deterministically the required values of these variables by, for example, modelling with systems of differential equations must fail when there is any difficulty in defining the systems criteria for success. In a similar way, the physical planner must renounce the aim of designing building systems on the basis of optimizing what amounts to a quite arbitrary utility function—whether this is in the location of facilities or anything else.

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Composition of Interstellar Grains

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A mixture of meteoritic silicate, silicon carbide, and graphite particles can explain all the observed properties of interstellar grains.

It has been suggested¹⁻⁴ that particles of magnesium silicate, aluminium silicate, silicon carbide, and carbon can be formed in the atmospheres of cool stars and expelled into interstellar space. Meteoritic silicate particles⁵ will also be formed in planetary systems. Observations in the far-infrared indicate the presence of circumstellar⁶, nebular⁷ and interstellar⁸ silicate particles. Recent observations⁹ seem to rule out significant amounts of water ice in interstellar space.

The observations which any "mixture" of interstellar grains should explain with the proper wavelength dependence are¹⁰: (1) the interstellar extinction; (2) the interstellar polarization; (3) the scattering by reflexion nebulae; and (4) the diffusely scattered galactic radiation. In view of the theoretical studies and observational evidence quoted above¹⁻⁹ we investigated whether a mixture of grains of graphite, silicon carbide, and meteoritic silicates in interstellar space can account for all these observations. We present here preliminary results of our computations showing that such a mixture indeed can account for all the observations.

If $n(r)dr$ is the fractional number of particles in the interval r and $r+dr$ then the cross-sections C_{ext} , C_{sca} , and C_{back} are given by^{11,12}

$$C_i(\lambda) = \int_0^\infty Q_i(r, \lambda) \pi r^2 n(r) dr \quad (1)$$

where Q_i is the efficiency parameter of the process involved. The phase parameter, g , is derived from

$$g C_{\text{sca}}(\lambda) = \int_0^\infty g Q_{\text{sca}}(r, \lambda) \pi r^2 n(r) dr \quad (2)$$

The size distribution function arbitrarily adopted is that given by van de Hulst for the Oort-van de Hulst size distribution (see ref. 12, page 70):

$$n(r)dr = e^{-0.69(r/\bar{r})^{2.8}} dr / \int_0^\infty e^{-0.69(r/\bar{r})^{2.8}} dr \quad (3)$$

where $\bar{r}$ is the radius such that $n(o)/n(\bar{r}) = 2$.

Complex refractive indices for graphite in the wavelength range $\lambda 6200 \text{ \AA}$ – $2480 \text{ \AA}$ were measured from Fig. 5 of Greenaway *et al.*¹³ for the case when the electric vector is perpendicular to the crystal axis. The values for $\lambda 6410 \text{ \AA}$ were extrapolated from their Fig. 5. The values for three wavelengths in the infrared were taken from Wickramasinghe¹². The values between $\lambda 2400 \text{ \AA}$ and $\lambda 1050 \text{ \AA}$ were measured from Fig. 2 of Carter *et al.*¹⁴. For meteoritic silicates Dorschner⁵ has suggested the following wavelength-independent values of

n , the real part of the refractive index, and k , the imaginary part: $n=1.70$, and $k=0.05$ to 0.30 . He points out that shortward of $3000 \text{ \AA}$, k will vary with wavelength. We adopted $n=1.70$ and $k=0.05$ for the whole wavelength range from $2.13 \mu\text{m}$ to $1050 \text{ \AA}$. A different value of k will not change our results significantly. For silicon carbide we used optical constants measured for the hexagonal form, with the electric vector perpendicular to the crystal axis. We measured n from Choyke and Patrick¹⁵ in the wavelength range $2.13 \mu\text{m}$ – $3360 \text{ \AA}$. In the wavelength range $3360 \text{ \AA}$ to $1240 \text{ \AA}$ it was measured from Philipp and Taft¹⁶, from which we also measured k . The value of k was taken to be zero for wavelengths longward of $3000 \text{ \AA}$. Friedemann³ has plotted n and k for SiC as a function of wavelength; he has also plotted Q_{ext} for several sizes.

Now the question arises how much each constituent contributes to a given phenomenon at a given wavelength λ . If we normalize such that $\Delta m = 0.0$ at $\lambda^{-1} = 1.83 \mu\text{m}^{-1}$ ($\lambda = 5565 \text{ \AA}$), and $\Delta m = 1.0$ at $\lambda^{-1} = 2.30 \mu\text{m}^{-1}$ ($\lambda = 4350 \text{ \AA}$), then the total extinction Δm^{tot} from the mixture at a wavelength λ can be written as

$$\Delta m^{\text{tot}}(\lambda^{-1}) = \frac{F_{\text{ext}}(\lambda^{-1}) - F_{\text{ext}}(1.83 \mu\text{m}^{-1})}{F_{\text{ext}}(2.30 \mu\text{m}^{-1}) - F_{\text{ext}}(1.83 \mu\text{m}^{-1})} \quad (4)$$

where

$$F_{\text{ext}}(\lambda^{-1}) = C_{\text{ext}}^{\text{Gr}}(\lambda^{-1}) + Y C_{\text{ext}}^{\text{Sil}}(\lambda^{-1}) + Z C_{\text{ext}}^{\text{SiC}}(\lambda^{-1}) \quad (5)$$

Superscripts "Gr", "Sil", and "SiC" refer to graphite, silicates and silicon carbide respectively, and Y and Z are respectively the number of silicate and silicon carbide grains relative to graphite grains. We can also calculate the normalized extinction for each constituent separately from

$$\Delta m^j(\lambda^{-1}) = \frac{C_{\text{ext}}^j(\lambda^{-1}) - C_{\text{ext}}^j(1.83 \mu\text{m}^{-1})}{C_{\text{ext}}^j(2.30 \mu\text{m}^{-1}) - C_{\text{ext}}^j(1.83 \mu\text{m}^{-1})} \quad (6)$$

where the j 's refer to the three constituents.

Another way of defining $\Delta m^{\text{tot}}(\lambda^{-1})$ is, following Hoyle and Wickramasinghe¹⁷, by writing that at $\lambda^{-1} = 2.30 \mu\text{m}^{-1}$, Y' times as much extinction is caused by silicates and Z' times as much by silicon carbide as by graphite. Relations between Y and Y' and Z and Z' can be easily obtained.

Interstellar Extinction

Recent observations of interstellar extinction in the ultraviolet by Stecher¹⁸ and Bless and Savage¹⁹ have confirmed the existence of a hump at $\lambda^{-1} \approx 4.6 \mu\text{m}^{-1}$ first reported by Stecher²⁰. This feature has been attributed to graphite^{21,22}. The observations of Stecher¹⁸ and the preliminary results obtained with the OAO-A2 satellite¹⁹ (these latter extinction curves have also been reproduced by Code²³, and the observations¹⁹ of ζ Oph and σ Sco are also indicated in our Fig. 4) can be summarized as follows: (1) all the extinction curves show a well pronounced, rather narrow, hump between $4.5 \mu\text{m}^{-1} \lesssim \lambda^{-1} \lesssim 4.7 \mu\text{m}^{-1}$; (2) all the extinction curves show a rather flat minimum between $5.5 \mu\text{m}^{-1} \lesssim \lambda^{-1} \lesssim 6.5 \mu\text{m}^{-1}$; (3) most of the curves show a rapid rise in the far ultraviolet;

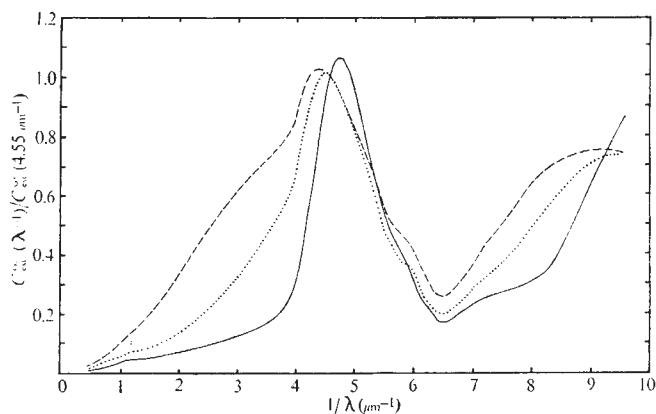


Fig. 1 The ratio of extinction cross-section to that at $4.55 \mu\text{m}^{-1}$ of graphite particles for three values of $\bar{r}_{Gr}$: —, $\bar{r}_{Gr} = 0.010 \mu\text{m}$; ···, $\bar{r}_{Gr} = 0.025 \mu\text{m}$; ---, $\bar{r}_{Gr} = 0.040 \mu\text{m}$.

(4) there seem to be real differences in the extinction curves, the differences being most pronounced in the far ultraviolet. The two stars ζ Oph and σ Sco shown in Fig. 4 represent about the extreme limit of the variations from star to star. Differences in the extinction curves also exist in the visual and infrared wavelength regions (Johnson²⁴). To get a good understanding of the distribution, composition, and size of the interstellar grains it is therefore advisable to consider the extinction curve for each star separately, over the entire available wavelength range, rather than a "mean" extinction curve.

Bless and Savage¹⁹ have compared their observations with a few recently published theoretical extinction curves. They conclude that none of the proposed models of interstellar grains gives a good fit to the observations, and suggest that the narrowness and the position of the hump at $\lambda^{-1} \approx 4.6 \mu\text{m}^{-1}$ indicate that it probably originates from small graphite particles. In Fig. 1 we have plotted the ratio $C_{ext}^{Gr}(\lambda^{-1})/C_{ext}^{Gr}(4.55 \mu\text{m}^{-1})$ for values of $\bar{r}_{Gr}$ of 0.010 μm , 0.025 μm and 0.040 μm . For $\bar{r}_{Gr} = 0.010 \mu\text{m}$ the maximum is at $\lambda^{-1} \approx 4.70 \mu\text{m}^{-1}$ and the hump is very narrow, but as we increase $\bar{r}_{Gr}$ the maximum shifts slowly towards longer wavelengths and the hump becomes broader. The minimum for all the curves is at $\lambda^{-1} = 6.45 \mu\text{m}^{-1}$. The curves rise again in the far ultraviolet but never to more than about $0.85 C_{ext}^{Gr}(4.55 \mu\text{m}^{-1})$. From a comparison of these curves with the observations it is clear that only small sized graphite particles can cause a well pronounced, rather narrow, hump. The small-sized graphite particles cannot give sufficient extinction in the wavelength region $\lambda^{-1} \lesssim 3.0 \mu\text{m}^{-1}$ if the extinction at the extinction maximum is fixed to the observational value. Thus we must have another constituent which gives us sufficient extinction in the wavelength range $\lambda^{-1} \lesssim 3.0 \mu\text{m}^{-1}$ but becomes "neutral" thereafter. Yet another constituent is needed to explain the rapidly rising extinction in the far ultraviolet.

We apparently need three different constituents to explain the entire extinction curve. Because in the infrared and visual region SiC has refractive index of about 2.7, with the imaginary part being very small, silicon carbide particles of reasonable size are a likely candidate for this region. Silicates have a refractive index of 1.70 and a small imaginary part in the entire wavelength range, so that silicate particles of reasonable size are candidates for the far ultraviolet region. We have plotted $C_{ext}^{SiC}(\lambda^{-1})$ in Fig. 2 and $\Delta m^{tot}(\lambda^{-1})$ in Fig. 3 for various values of $\bar{r}_{SiC}$ and $\bar{r}_{SiI}$. The principal features of these curves will persist with any change in the form of the size distribution function as long as the range of sizes is sufficiently large (see also refs. 2, 3, 11, 17 and 25). In the curves for silicon carbide there is a broad dip with the minimum at $\lambda^{-1} \approx 5.3 \mu\text{m}^{-1}$. This is because of the onset of strong absorption arising from direct transitions between conduction and valence bands in SiC. This feature is very clearly present in its albedo curve.

Figs. 1–3 demonstrate that the general features of the interstellar extinction curves can be reproduced with a mixture of graphite particles having $0.010 \mu\text{m} \lesssim \bar{r}_{Gr} \lesssim 0.025 \mu\text{m}$, SiC particles having $\bar{r}_{SiC} \approx 0.075 \mu\text{m}$, and silicate particles having $\bar{r}_{SiI}$ between approximately $0.030 \mu\text{m}$ and $0.045 \mu\text{m}$. For a detailed fit it is very important that we have accurate and carefully reduced observations, accurate refractive indices, and more information about the probable forms of the size distribution (this will probably be different for different constituents). We shall now consider situations which will not be appreciably altered by any likely changes in the above three factors.

In Fig. 4 we plot the total normalized extinction $\Delta m^{tot}(\lambda^{-1})$ for the mixture, using equation (4) for $\bar{r}_{Gr} = 0.025 \mu\text{m}$, $\bar{r}_{SiI} = 0.045 \mu\text{m}$, $\bar{r}_{SiC} = 0.075 \mu\text{m}$, $Y' = 0.8$, and $Z' = 1.8$. The filled circles represent the mean observational values found by Boggess and Borgman²⁶. The OAO-A2 observations of the stars ζ Oph and σ Sco are also indicated. Refractive indices for SiC are available in the ultraviolet only down to $\lambda 1240 \text{ \AA}$. Its extinction curve does not seem to vary much in this wavelength range, so we have assumed $C_{ext}^{SiC}(\lambda < 1240 \text{ \AA}) = C_{ext}^{SiC}(\lambda = 1240 \text{ \AA})$. We have also plotted the individual contributions from the three constituents. A comparison with the observations of Stecher¹⁸ and Bless and Savage¹⁹ indicates that the agreement is reasonable. From Fig. 4 we see that the contribution from SiC has a flat maximum at $\lambda^{-1} \approx 2.5 \mu\text{m}^{-1}$ and decreases slowly thereafter, causing a significant change in the slope of the extinction curve starting at slightly smaller values of λ^{-1} . It is well known^{27,28} that there is a distinct change in the slope of the extinction curve at $\lambda^{-1} \approx 2.3 \mu\text{m}^{-1}$. We suggest that this "kink" in the observed extinction curves is caused by SiC particles with a size distribution such that the maximum in their extinction curve is at $\lambda^{-1} \approx 2.5 \mu\text{m}^{-1}$. It is very interesting to note that a "kink" at about these wavelengths has also been observed²⁹ in the extinction curves of a few heavily reddened stars in the large Magellanic cloud, suggesting that in the LMC also there are at least two different constituents of the interstellar grains.

To illustrate some effects caused by small changes in the size of particles and in the relative contributions from the three constituents, we have plotted in Fig. 5 theoretical extinction curves with the following parameters: *a*, $\bar{r}_{Gr} = 0.020 \mu\text{m}$, $\bar{r}_{SiI} = 0.045 \mu\text{m}$, $\bar{r}_{SiC} = 0.075 \mu\text{m}$, $Y' = 2.0$, and $Z' = 3.5$; *b*, $\bar{r}_{Gr} = 0.015 \mu\text{m}$, $\bar{r}_{SiI} = 0.045 \mu\text{m}$, $\bar{r}_{SiC} = 0.070 \mu\text{m}$, $Y' = 3.0$, and $Z' = 10.0$; and *c*, $\bar{r}_{Gr} = 0.015 \mu\text{m}$, $\bar{r}_{SiI} = 0.030 \mu\text{m}$, $\bar{r}_{SiC} = 0.075 \mu\text{m}$, $Y' = 1.2$, and $Z' = 8.0$. In curve (*a*) the graphite and silicon carbide particles are slightly larger and the relative contribution to the extinction from silicates and SiC is much less. The "kink" is present at $\lambda^{-1} \approx 2.3 \mu\text{m}^{-1}$ in all the curves, but the subsequent slopes are different. Nandy²⁸ finds different slopes in Cygnus and Perseus which in our opinion need such

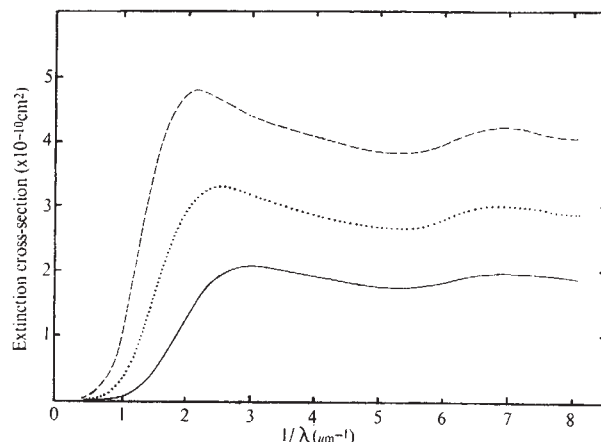


Fig. 2 Extinction cross-sections of silicon carbide particles for three values of $\bar{r}_{SiC}$: ---, $0.090 \mu\text{m}$; ···, $0.075 \mu\text{m}$; —, $0.060 \mu\text{m}$.

an explanation. We are, however, not attempting any detailed fits at this stage.

As we see from Fig. 4, silicates are the chief contributors to the rapidly rising extinction in the far ultraviolet. The rapidly increasing extinction from silicates in this wavelength range shifts considerably the graphite minimum at $\lambda^{-1} \approx 6.54 \mu\text{m}^{-1}$ to a smaller value of λ^{-1} and shifts slightly the graphite peak to a larger value of λ^{-1} . The exact amounts of these shifts depend primarily on $\bar{r}_{\text{SiH}}$ and not on Y' , the relative contribution from silicates. For smaller values of $\bar{r}_{\text{SiH}}$ the extinction from silicates increases much more rapidly, and we see from curve (c) in Fig. 5 that the total extinction rises very rapidly in the far ultraviolet; the minimum is at $\lambda^{-1} \approx 5.5 \mu\text{m}^{-1}$, and the peak is at $\lambda^{-1} \approx 4.7 \mu\text{m}^{-1}$. The far ultraviolet extinction curves for $\zeta \text{ Per}^{18,19}$ and $\zeta \text{ Oph}^{19}$ (Fig. 4) look somewhat like this.

We have not taken into account the refractive indices for E_{ie} , the electric vector parallel to the crystal axis. Greenaway *et al.* have obtained for this orientation for the wavelength range quoted above values of $n \approx 1.54$ and $k = 0.00$ almost independently of wavelength. They suggest that there is structure in the values of the refractive indices for $\lambda^{-1} \gtrsim 4.0 \mu\text{m}^{-1}$ but the values are not known. The component of extinction arising from light with E_{ie} will not be negligible in the ultraviolet²¹. The exact contribution cannot be calculated because the theory of scattering by particles having anisotropic optical properties is not available²⁵. Thus we cannot say how the shape of the extinction curve will be actually affected if we have to include this contribution.

Interstellar Polarization

There seem to be variations in the wavelength dependence of interstellar polarization^{30,31} from star to star. But in the mean curve given by Coyne and Gehrels³⁰ there is a weak maximum at $\lambda^{-1} \approx 1.9 \mu\text{m}^{-1}$. The observations so far cover the wavelength range $1 \mu\text{m}$ – $0.3 \mu\text{m}$. Because in our model SiC is the chief contributor to the extinction in this wavelength range, it should also be the chief contributor to polarization. Hexagonal SiC is a birefringent crystal. The difference³² in its ordinary and extraordinary refractive indices is about 0.05 in the wavelength range for which the experimental results are available, $0.71 \mu\text{m}$ – $0.43 \mu\text{m}$, the ordinary refractive index being bigger. The Q_{H} curve, which is calculated with the extraordinary refractive index, will therefore be shifted slightly towards higher values of λ^{-1} relative to the Q_{E} curve, which is calculated with the ordinary refractive index. At a given wavelength the difference $Q_{\text{E}} - Q_{\text{H}}$ integrated over the size distribution will give us a measure of polarization. Any anisotropy in the shape of the particles will increase the polarization²⁵ because the refractive index is so high. Because the maximum in the extinction curve from SiC is required to

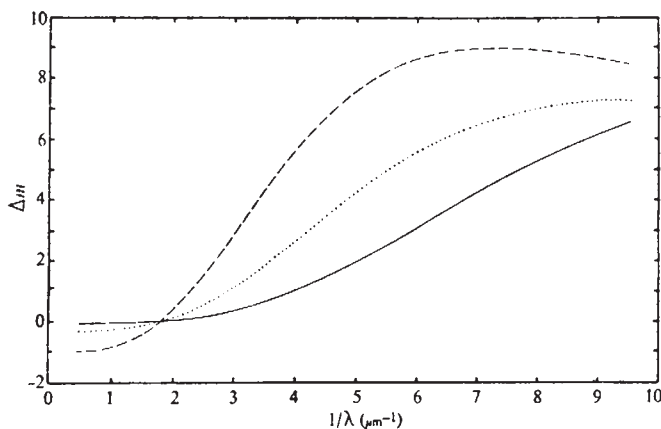


Fig. 3 Normalized extinction using equation (6) for silicate particles for three values of $\bar{r}_{\text{SiH}}$: ---, $0.060 \mu\text{m}$; ..., $0.045 \mu\text{m}$; —, $0.030 \mu\text{m}$. The actual values have been multiplied by the factors 1 (---), $\frac{1}{2}$ (...) or 0.1 (—).

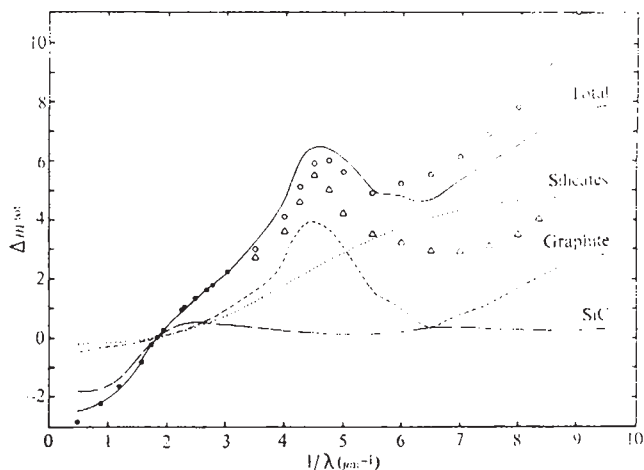


Fig. 4 A theoretical extinction curve (solid line) using equation (4) for $\bar{r}_{\text{SiH}} = 0.045 \mu\text{m}$, $\bar{r}_{\text{Gr}} = 0.025 \mu\text{m}$, $\bar{r}_{\text{SiC}} = 0.075 \mu\text{m}$, $Y' = 0.8$, $Z' = 1.8$, $Y = 0.374$ and $Z = 0.060$. Individual contributions of the three constituents are indicated. OAO-A2 observations¹⁹ of two stars, $\zeta \text{ Oph}$ (○) and $\sigma \text{ Sco}$ (△), and the mean observational values found by Boggess and Borgman²⁶ (●) are also shown.

occur at about $\lambda^{-1} \approx 2.5 \mu\text{m}^{-1}$, the maximum in its polarization curve will occur at a smaller value of λ^{-1} (refs. 11, 25). We therefore suggest that the observed "kink" at $\lambda^{-1} \approx 2.3 \mu\text{m}^{-1}$ in the interstellar extinction curve and the observed maximum in the interstellar polarization curve at $\lambda^{-1} \approx 1.9 \mu\text{m}^{-1}$ have the same origin: in the mixture of grains of SiC, graphite, and silicates the size distribution of SiC particles is such that in its extinction curve the maximum is at $\lambda^{-1} \approx 2.5 \mu\text{m}^{-1}$.

To explain the circumstellar polarization Friedemann³ has suggested that SiC particles can be aligned in the stellar magnetic field, but the problem of alignment for both the circumstellar and interstellar cases has yet to be investigated quantitatively.

Regions containing Young, Hot Stars

An important test any mixture of interstellar grains should satisfy is whether the observed "anomalous" behaviour of interstellar extinction (refs. 19, 24, 34–36 and private communication from R. C. Bless and B. D. Savage) and interstellar polarization^{31,37,38} in the regions containing young, hot stars can be explained without additional assumptions. In brief, the observations of stars in these regions as compared with stars in other regions indicate: (1) higher values of R , the ratio of total to selective extinction; (2) marked differences in the shapes of the extinction and polarization curves; (3) the maximum in the polarization curves at much smaller values of λ^{-1} resulting in higher values of P^V/P^R , the ratio of polarizations; (4) lower values of extinction throughout the ultraviolet ($\sigma \text{ Sco}^{19}$, Fig. 4), with very low values in the far ultraviolet ($\sigma \text{ Sco}^{19}$, $\theta \text{ Orionis}^{35,36}$, and, possibly, 42 Orionis^{36}). If we assume³⁵ that Δm^{tot} at $1270 \text{ \AA}$ is 8.0, we find $\sigma \text{ Sco}$ is brighter by 1.7 magnitudes at this wavelength. OAO-A2 observations also indicate very high values of flux or very low extinction for the Trapezium stars (private communication from R. C. Bless and B. D. Savage).

Code (manuscript in preparation) has suggested that the angular resolution of the instruments working in the rocket ultraviolet is not good enough to separate the nebulosity associated with $\sigma \text{ Sco}$ and $\theta \text{ Orionis}$ from the stellar contribution. Corrections must be made for the contribution of the nebulosity. His preliminary calculations show that the nebular contribution does not seem to be sufficient to account for the large apparent increase in the brightness, thus indicating less extinction.

It has been suggested (refs. 24, 31, 34, 35, 37–39 and A. D. Code, manuscript in preparation) that these observations can

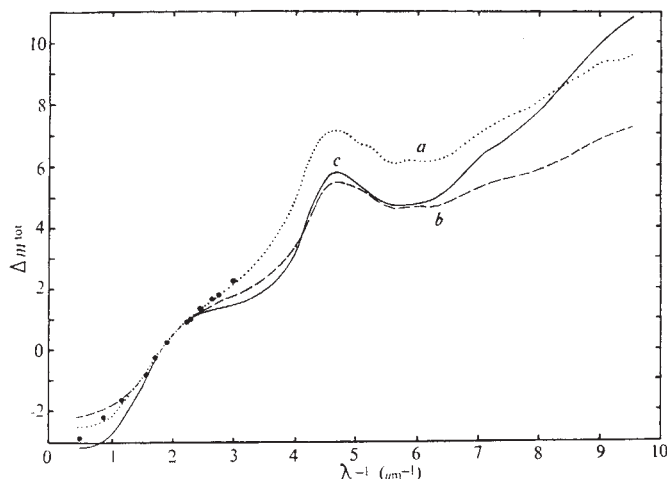


Fig. 5 Three theoretical extinction curves using equation (4) for: *a*, $\bar{r}_{\text{SiC}}=0.045 \mu\text{m}$, $\bar{r}_{\text{Gr}}=0.020 \mu\text{m}$, $\bar{r}_{\text{SiC}}=0.075 \mu\text{m}$, $Y'=2.0$, $Z'=3.5$, $Y=0.344$ and $Z=0.044$; *b*, $\bar{r}_{\text{SiC}}=0.045 \mu\text{m}$, $\bar{r}_{\text{Gr}}=0.015 \mu\text{m}$, $\bar{r}_{\text{SiC}}=0.070 \mu\text{m}$, $Y'=3.0$, $Z'=10.0$, $Y=0.159$ and $Z=0.035$; *c*, $\bar{r}_{\text{SiC}}=0.030 \mu\text{m}$, $\bar{r}_{\text{Gr}}=0.015 \mu\text{m}$, $\bar{r}_{\text{SiC}}=0.075 \mu\text{m}$, $Y'=1.2$, $Z'=8.0$, $Y=0.508$ and $Z=0.031$. ●, Boggess and Borgman²⁶.

be explained in terms of larger particles. The argument is that in the neighbourhood of young, hot stars the size distribution of the particles will be modified such that there is a sharp decline in the numbers of smaller particles. If we selectively remove smaller size particles from our model, we see that: (1) the maximum in the extinction curve due to SiC will be at values of λ^{-1} smaller than $2.5 \mu\text{m}^{-1}$ (ref. 25), and consequently the "kink" in the observed extinction curve will be at a smaller value of λ^{-1} , the value of R will be larger, and the maximum in the polarization curve will also be at a smaller value of λ^{-1} ; (2) the extinction in the far ultraviolet arising from silicates will tend to become neutral so the normalized extinction from the mixture will not show a rapid rise in this wavelength range; (3) the maximum in the normalized curve will have a lower value, will be at $\lambda^{-1} \approx 4.5 \mu\text{m}^{-1}$ or less, and will be somewhat broader. All these predictions are in excellent qualitative agreement with observations.

It must be kept in mind that the "anomalies" will become pronounced only if the "local" component where the size distribution of the particles has been modified is sizable in comparison with the "interstellar" component.

It seems that a good match to all the observations can be obtained with a mixture of grains of SiC, graphite, and silicates. The position of the "kink" in the extinction curve, the value of R , and the position of the maximum in the polarization curve will give us the first approximation to the size distribution of SiC particles. The exact position and width of the extinction maximum at about $\lambda^{-1} \approx 4.6 \mu\text{m}^{-1}$ and Δm^{tot} at this wavelength, the position and Δm^{tot} of the extinction minimum, and Δm^{tot} at some wavelength in the far ultraviolet, say $\lambda^{-1} = 9.0 \mu\text{m}^{-1}$, will give us the first approximation to the size distribution of silicate and graphite particles.

Reflexion Nebulae and Diffuse Galactic Radiation

With the parameters of Figs. 4 and 5, excellent agreement has been obtained for the back-scattering observations⁴⁰ of some reflexion nebulae in the Pleiades cluster.

In Fig. 6 we have plotted albedo, $\langle \gamma(\lambda^{-1}) \rangle$, and phase parameter, $\langle g(\lambda^{-1}) \rangle$, using equations of Wickramasinghe³³, for the parameters of Fig. 4 and Fig. 5b. A minor discontinuity at $\lambda^{-1} = 3.33 \mu\text{m}^{-1}$ arises from our assumption that k for SiC is zero for $\lambda^{-1} \leq 3.33 \mu\text{m}^{-1}$. Because k is not quite zero in this wavelength range, the actual value of the albedo will be slightly less for this wavelength range. In the visual

region the value of the albedo is also quite sensitive to the relative concentration of the SiC particles. From these curves we get, for $\lambda^{-1} \approx 2.5 \mu\text{m}^{-1}$, albedo ≈ 0.8 and $\langle g \rangle \approx 0.25$. These values are in good agreement with the observations of the diffuse galactic light in the B and U filters as interpreted by van de Hulst and de Jong¹⁰. For $\langle g \rangle = 0.25$ the albedo from their Table 4 is about 0.65 for both the filters. In Fig. 6 the maximum at $\approx 1.5 \mu\text{m}^{-1}$ in the value of $\langle g \rangle$ is due to SiC. Its position depends on the size distribution of SiC grains. A broad dip in the albedo curve with the minimum at $\lambda^{-1} \approx 4.6 \mu\text{m}^{-1}$ is due to SiC which has a minimum at about $4.7 \mu\text{m}^{-1}$ in its albedo. The observations⁴¹ of the diffuse ultraviolet radiation indicate a minimum in the albedo of interstellar grains at $\lambda^{-1} \approx 4.0 \mu\text{m}^{-1}$.

The observations of the diffuse far ultraviolet radiation (1350–1480 Å) and their interpretation⁴² indicate an albedo close to unity and $\langle g \rangle \approx 0.6$. The corresponding values from Fig. 6 are, $\langle \gamma \rangle \approx 0.7$ and $\langle g \rangle \approx 0.6$, indicating reasonable agreement. The correct refractive indices for the silicates in the far ultraviolet are not known, so that their effect on the values of albedo in this wavelength range cannot be estimated.

Abundances

From the parameters of Figs. 4 and 5 relative masses of the three constituents are roughly: for 1 g of graphite grains there are about 4 g of SiC grains and 5 g of silicate grains. To explain 1 mag/kpc of extinction at $\lambda = 5565 \text{ Å}$ we need about $8 \times 10^{-27} \text{ g cm}^{-3}$ in the form of dust. On comparing the abundances of elements we need with those quoted by Greenberg²⁵, and assuming 1 H atom cm^{-3} in the interstellar space, we find that all the elements except Si are in adequate supply. We need about $3 \times 10^{-27} \text{ g cm}^{-3}$ of Si whereas only about $1.2 \times 10^{-27} \text{ g cm}^{-3}$ seems to be available in interstellar space. In the form of size distribution we have adopted, most of the mass is from smaller particles and most of the extinction from larger particles. Especially in the case of SiC we do not need the smaller particles which are contributing to the extinction in the ultraviolet in its extinction curve. Friedemann³ has pointed out that smaller SiC particles will not be formed. Thus the amount of SiC, and therefore of Si, needed will be substantially reduced. But according to this model it seems likely that most of the Si in interstellar space is tied in the grains.

SiC particles will be formed in carbon-rich stars^{2–4} and will be the chief condensate in stars having C/O ratio approximately unity⁴. In oxygen-rich stars they may acquire mantles of SiO_2 (refs. 2, 3). No estimate is available about the relative abundances of graphite, SiC and silicates, and we cannot say whether the amount of SiC needed is excessive.

The fundamental absorption band in SiC^{43} is at $12.6 \mu\text{m}$. Mie calculations show an absorption band at this wavelength but it is completely dwarfed by an absorption feature at $\sim 10.75 \mu\text{m}$, where $n^2 - k^2$ equals -2 . This feature will become broad if the particles are not exactly spherical. Kumar (private

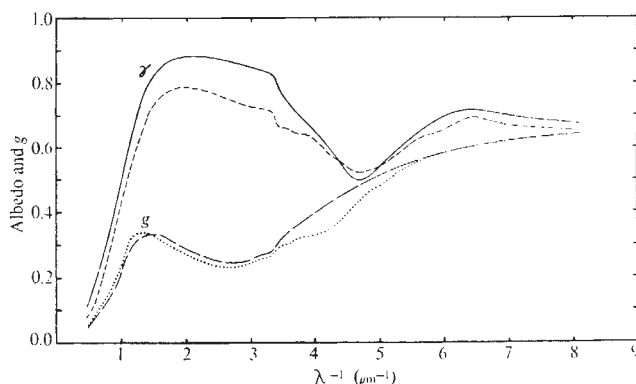


Fig. 6 Theoretical curves for albedo and g , the phase parameter (I) for the parameters of Fig. 4, albedo (---) and g (---); (II) for the parameters of Fig. 5b, albedo (—) and g (—).

communication) has suggested transitions in SiC containing impurities can explain the observed diffuse interstellar absorption features.

It seems that all the observed phenomena in which interstellar particles are participating can be explained very well with a mixture of grains of graphite, silicon carbide, and meteoritic silicates, but detailed quantitative investigation is needed. Optical properties of these substances, with and without impurities, should be investigated in detail in the laboratory. More theoretical work is needed to minimize the arbitrariness with which the size distribution of particles is assumed. Observations of variations in the intrinsic polarization of the late type variable stars where grain formation is taking place will also help in our understanding of the size and composition of particles.

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Proposed Mechanism for the Biological Assembly of Collagen Triple Helix

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If the three polypeptide chains of collagen had terminal segments with structures which facilitated rapid association and were then discarded, a puzzle might be solved. Such a mechanism could explain why biosynthesis of collagen takes minutes *in vivo* but much longer *in vitro*.

TROPOCOLLAGEN (TC) consists of three protein chains with similar, specific and very characteristic amino-acid compositions, twisted into a plectonemic¹ triple helix of about 100

turns. *In vivo*, the triple helix seems to be assembled in minutes, whereas *in vitro*, a solution of the separated chains must be annealed with the concentration and temperature carefully chosen, to ensure that each helix is composed of the appropriate chains in register. Assembly may take several days and the yield is low. To account for this difference, I propose that the protein, chains when first synthesized, are longer than those in TC, and that the peptide extensions aggregate rapidly and specifically, so that three appropriate α chains are brought together to allow the rest of the molecule to coil into the in-register triple helix, and that the peptide extensions are removed later enzymatically.

Structure and Biosynthesis

TC usually consists of two identical, $\alpha 1$, chains, and a third different, $\alpha 2$, chain^{2,3}. The molecular weight of the chains is

approximately 100,000; they have specific amino-acid sequences without any repeating peptides. Every third residue in the chain is glycine, except in short "non-collagenous" telopeptides⁴ at the N-terminal ends of the chains. The collagenous region is remarkably constant in amino-acid sequence when avian and mammalian species are compared: there is more variation between species in the telopeptide regions⁵.

The triple helix structure is stabilized by a large proportion of proline and hydroxyproline residues, and by hydrogen bonds between the chains. The low solubility of connective tissue in acid or neutral buffers is accounted for by covalent crosslinks between the chains within the TC molecule or between the chains of TC molecules which are adjacent in the collagen fibril. Both possible forms of the β component, in which two α chains are linked, and all four of the γ component, in which three are linked, have been demonstrated in solutions of denatured collagen⁶⁻⁸.

In collagen fibrils, adjacent TC molecules (length approximately 2900 Å) are staggered with respect to one another by a fraction of their length equal to 1, 2, 3, or 4 divided by 4.4. Along the axis of the fibril there are gaps between in-line TC molecules equal to 13% of the length of the molecule⁹.

The biosynthesis of collagen has already been discussed¹⁰. Attempts to isolate collagen-forming polyribosomal aggregates and determine the number of ribosomes have given equivocal results, so that it has not been possible to infer whether single α chains or proteins three times the length of α chains which are converted later by hydrolytic fission to α chains are synthesized at the polysome¹¹. Perhaps the fact that two of the α chains are identical makes it more likely that the polysomes are the site of synthesis of α chains, not complete TC molecules.

Assembly of Triple Helix

Many experiments have shown that enzyme or enzyme subunit proteins can be denatured to a random configuration and then, if denaturing conditions are replaced by physiological conditions, the protein chains assume again the specific native configuration. Furthermore, if an enzyme is composed of, say, two or four subunits, the renatured subunits assemble into active enzyme molecules^{12,13}. Collagen is an exception to this general scheme (insulin is another). When mammalian tropocollagen is dissolved in a physiological buffer, the triple helix can be made to unwind and the chains separate, unless they are cross-linked by increasing the temperature above 37° C, the physiological temperature of the animal. If a denatured TC solution is cooled to 4° C triple helical sections rapidly form, but the α chains are out of register and not necessarily $\alpha_1 \alpha_1 \alpha_2$; at 25° C, more triple helices are formed from two α_1 and one α_2 chains in register, but more slowly. For example, in one experiment 50% renaturation was achieved in 50 h at 25° C. These experiments imply that below the denaturation temperature, random assemblies of α chains, out of register but with some triple helical sections, are less stable than the in-register $\alpha_1 \alpha_1 \alpha_2$ triple helix. Conversion to the more stable form is slow, however^{3,14-16}.

These results contrast with the observation that in tissue culture and *in vivo*, radioactive proline appears, as hydroxyproline, in TC within minutes of being added to the system¹⁷.

Proposed Mechanism

My suggestion, which is illustrated in Fig. 1, is that α chains, when just completed at the polyribosomal aggregate, are longer than those in soluble TC. These extensions to the α chains interact, two α_1 extensions and one α_2 extension, to form a stable assembly. Thus the correct chains are brought together in register. The interaction between these extensions then allows the formation of the triple helix in register, so that "registration peptides" might be an appropriate name. Regis-

tration peptides (RPs) may be continuations of the telopeptides at the N-terminal ends of the α chains which, because proteins are synthesized beginning at their N-termini, would allow registration before synthesis of the chains was complete; alternatively, attachment of the RPs to the C-termini would postpone registration until the chains were completed.

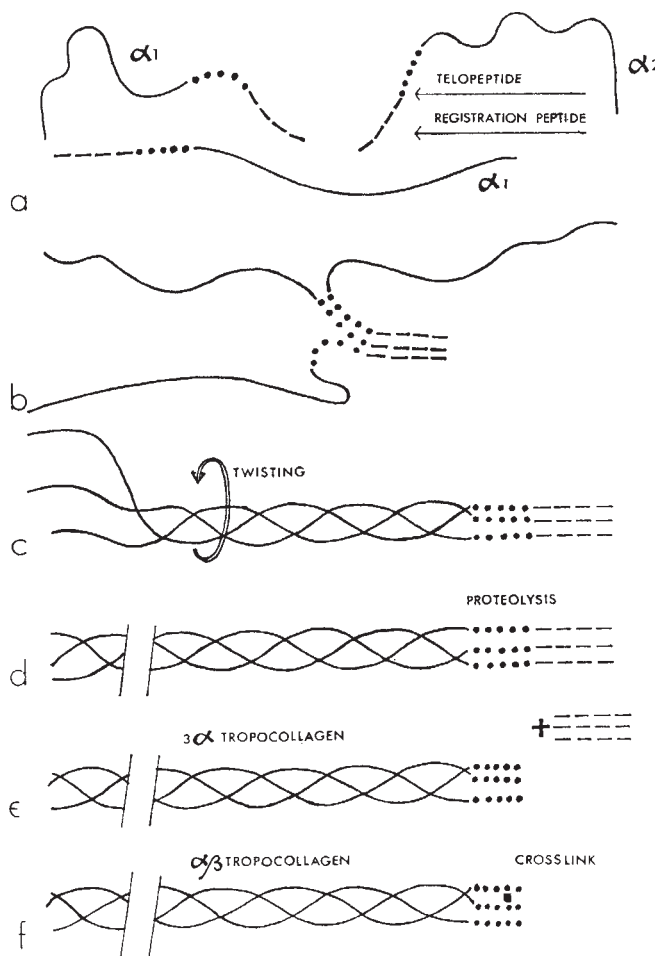


Fig. 1 *a* to *e*, Stages in the mechanism proposed for the biological assembly of the tropocollagen triple helix. The theory postulates that each new α chain leaves the polyribosomal aggregate with a registration peptide attached to one end. In *a* they are shown, arbitrarily, at the N-terminal, telopeptide ends of the chains. Registration peptides from two α_1 and one α_2 chain interact laterally and specifically to form a stable aggregate (*b*). This enables the collagenous body of the molecule to twist (*c*) into an in-register triple helix (*d*) composed of the correct α chains. The discontinuity is to indicate that each α chain has 100 turns in the tropocollagen molecule. The registration peptide lateral aggregate is then removed enzymatically (*e*). Crosslinkages can then form within (*f*) the tropocollagen molecule, or between adjacent tropocollagen molecules in the fibril.

TC without RPs can form collagen fibrils *in vitro* and telopeptides seem to facilitate assembly^{3,4,9}. I propose, however, that the triple helix, with RPs still attached, becomes part of the fibril. Then the structure is stabilized by secondary valence forces—hydrogen bonds, ionic interactions, hydrophobic bonds between adjacent TC molecules, all mediated by amino-acid side chains. It may be further stabilized by intra-TC or inter-TC covalent bonds. During or after stabilization of the structure in this way, the RPs must be removed proteolytically to leave the usual α_1 and α_2 chains. No suggestion is made

about the size and shape of the specific aggregate of RPs, but a hole about 400 Å long and 20 Å in diameter is available between the ends of in-line TC molecules in the fibril, to accommodate it⁹. An attractive possibility is that the individual RPs, unlike the collagenous body of the molecule and the telopeptide region⁴, have a high proportion of amino-acid residues which favour the (keratin) α -helix configuration. In the α -helical regions of myosin¹⁸ and keratin¹⁹, the helices seem to have aggregated laterally, specifically.

The proposed mechanism is clearly analogous to the conversion of zymogens into active enzymes²⁰, the reactions of prothrombin and fibrinogen in the clotting of blood²⁰, and to the change from the single chain proinsulin into insulin with two protein chains²¹, all of which occur in nature by the excision of peptides from the original molecules.

Registration Peptides

The mechanism implies that at the physiological temperature of the animal, the specific aggregate formed from the RP extensions of the three correct α chains is stable, whereas the triple helix formed from the collagenous body of the molecule (with the attached telopeptides) is in equilibrium with its denatured form. This may have advantages in growth and development and in regeneration and wound healing. If rapid metabolic turnover is necessary, removal of the RPs would make it possible for the three chains to separate; alternatively, if metabolic stability and mechanical strength are needed, these can be achieved by cross-linking within and between the TC molecules.

I propose two methods for the preparation of TC with registration peptides attached. The theory predicts that in collagen-synthesizing tissues there will be a small, steady state concentration of α chains with RPs attached, as precursors of collagen fibrils. (Indeed, Fessler and Smith²², using denatured collagen from chick fibroblasts in tissue culture, have detected protein with some properties of α chains but higher molecular weights, by gel filtration and gel electrophoresis.) Therefore, a TC preparation might be expected to contain a small proportion of molecules with the RPs attached (TC_{rp}). The theory suggests ways in which TC_{rp} might be separated from TC, and certain properties of TC_{rp} which could be tested experimentally.

TC_{rp} would be more resistant to chain separation than 3 α TC or α BTTC. Therefore, in gel chromatography of denatured collagen²³, TC_{rp} might be eluted with γ TC. The chains in γ TC, however, are crosslinked covalently, whereas those in TC_{rp} are not, so that these forms might be separated by gel chromatography in more severe denaturing conditions, when TC_{rp}, but not γ TC, would separate into α chains with RPs attached.

Another property of α chains with RPs attached which might be used to separate them from other components of denatured collagen would be the expected ease with which they would renature to form TC_{rp}. In a method similar to that used to prepare γ TC (ref. 24), the temperature of a solution of mammalian collagen might be raised to 40° C, which would separate the chains of 3 α TC and α BTTC. Although the molecule is denatured and the chains untwisted, the chains of γ TC remain covalently crosslinked. The RPs of TC_{rp} might or might not separate at this temperature. As in the preparation of γ TC, lowering the temperature to 25° C would cause rapid reformation of the triple helix from γ TC, and would be expected to reform TC_{rp} whether or not the α chains with RPs attached had separated at 40° C. The reformed triple helices might then be separated by their ability to form collagen fibrils in salt solution, or lateral aggregates in register (SLS (ref. 9)) on the addition of ATP. As in the previous preparation method, it would be necessary to use the absence of covalent crosslinks in TC_{rp} to separate it from γ TC.

The properties of tropocollagen with registration peptides attached can be predicted. The RP theory suggests that there would be small differences in such physical properties as sedi-

mentation coefficient, intrinsic viscosity, optical rotatory dispersion and molecular weight, compared with TC, and differences in amino-acid composition and electrophoretic behaviour. Similar differences would be expected between α chains with and without RPs.

In a radioactive pulse labelling experiment the theory predicts a higher specific activity in TC_{rp} than in soluble TC as a whole. Similarly, in a pulse-chase experiment, the pulse should be chased from TC_{rp} into TC molecules without RPs. Pulse experiments²⁵ might also be used to discover whether RPs are C- or N-terminal.

Treatment with proteolytic enzymes would be expected to remove the RPs. This suggests that in attempting to prepare TC enriched with TC_{rp}, precautions should be taken to exclude or inhibit proteolytic activity. It also suggests why TC_{rp} may not previously have been noticed in collagen preparations, apart from Fessler and Smith's²² report. But a hydroxyproline-containing protein, which can only be denatured to α chains if first treated with pepsin, has been reported²⁶ when fibroblasts are cultured in the presence of an inhibitor of covalent cross-linking.

If the RP theory can be confirmed by further experiment, then a defect in the RP excision enzyme might be considered as a cause of some connective tissue diseases.

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Menstrual Synchrony and Suppression

by

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Synchrony and suppression among a group of women living together in a college dormitory suggest that social interaction can have a strong effect on the menstrual cycle.

STUDIES of the influence of pheromones on the oestrous cycles of mice¹⁻⁴, and of crowding on variables such as adrenalin production in mice and other species⁵ have suggested that social grouping can influence the balance of the endocrine system. Although there has been little direct investigation with humans, anecdotal and indirect observations have indicated that social groupings influence some aspects of the menstrual cycle. Menstrual synchrony is often reported by all-female living groups and by mothers, daughters and sisters who are living together. For example, the distribution of onsets of seven female lifeguards was scattered at the beginning of the summer, but after 3 months spent together the onset of all seven cycles fell within a 4 day period.

Indirect support is given by the investigation of Collet *et al.*⁶ on the effect of age on menstrual cycle patterning. A higher percentage of anovulatory cycles were reported for college age women than for older women. Although Collet *et al.* attributed this to a maturational factor, it is interesting that most of the college aged women attended all female schools. Considering the parallel with the Lee-Boot effect in mice¹ (groups consisting only of females become pseudopregnant or anoestrous), it seems possible that an interpersonal factor is operating together with the maturational factor.

Subjects were 135 females aged 17-22 yr—all residents of a dormitory in a suburban women's college. The dormitory in which they resided has four main corridors each with approximately twenty-five girls living in single and double rooms. Six smaller living areas, separated from the main corridors by at least one door, each house approximately eight girls in single rooms.

Three times during the academic year, each subject was asked when her last and second to last menstrual periods had begun; thus the date of onset was determined for all cycles between late September and early April. The average duration of menstruation and presence of dysmenorrhoea were noted. In addition, subjects estimated how many times each week they were in the company of males and listed by room number the girls ($N \leq 10$) with whom they spent the most time, indicating which two of these they saw most often.

The date of menstrual onset was compared for room mates and closest friends, for close friend groups and for living groups. Two people qualified as "closest friends" only if both had indicated that they saw each other most often. While menstrual cycle timing in women using birth control pills is individually invariant, these women were still included in the analysis, because their influence on the menstrual cycles of the others was unknown. For room mates and closest friends, the difference between the date of onset in October for one arbitrarily chosen member of the pair and the closest date of onset for the other was calculated. This difference was compared with a difference for March calculated in a similar way, but with one change: instead of choosing the closest onset dates for the pair, both onsets for March were chosen to follow

the initial October onset by an equal number of cycles. For example, if onset 6 occurred on March 10 for the first member of the pair, and onsets 5 and 6 for the other member occurred on March 1 and March 29 respectively, then the March 10 and March 29 dates were used to calculate the difference in onset. This procedure was used to minimize chance coincidences that did not result from a trend towards synchrony.

The Wilcoxon matched-pairs signed-ranks test⁷ was used to test for a significant decrease in the difference between onset dates of room mates and closest friends. This test utilizes both the direction and magnitude of change in differences and is therefore a relatively powerful test.

There was a significant increase in synchronization (that is, a decrease in the difference between onset dates) among room mates ($P \leq 0.0007$), among closest friends ($P \leq 0.003$) and among room mates and closest friends combined ($P \leq 0.0003$). The increase in synchrony for room mates did not differ significantly from the increase for closest friends. The increase in synchrony was further substantiated by non-overlapping confidence intervals, calculated for the median difference on onset dates⁸ (Table 1).

This synchrony might be due to some factor other than time spent with an individual; Koford⁹ has attributed synchrony of the breeding season in *Macaca mulata* on Cayo Santiago to common seasonal changes in available food. The fact that the subjects generally eat as a dormitory group in a common dining room might be a significant factor in creating synchrony. A similar life pattern and common, repeated stress periods might also effect synchrony. Subjects were therefore randomly paired and tested for synchrony within the dormitory as a whole, but no significant trend (N.S., $P \leq 0.8$) was found, and the confidence intervals for the median difference in onset date overlapped completely.

Group synchrony was also investigated and the data were analysed to verify that the decrease in difference between onset dates was a true measure of synchrony. All subjects were divided into fifteen groups of close friends ($5 \leq N \leq 10$), using the lists of close friends made by each subject. During the interview, it was stressed to each subject that her list of "close friends" should include the people she saw most often and with whom she spent the most time, not necessarily those with whom she felt the closest. But because there is usually some overlap, the term "close friends" was adopted. Only subjects who mutually listed each other were included in a group.

A mean onset date (μ_i) was determined for each group in October, late November, January, late February and April. As before, the onset dates (X_i) being compared, each followed the October onset (X_1) by an equal number of cycles. The mean individual difference from the group onset mean

$$\frac{\sum (X_i - \mu_i)}{n}$$

was determined for each group and compared across time in two ways. First, a linear rank method, designed by Page¹⁰ to

Table 1 Confidence Intervals (> 0.99 , in days) for the Median Difference in Onset Date between Members of the Pair

	October	March
Close friends and room mates N=66	7 < M < 10	3 < M < 7
Random pairs N=33	6 < M < 14	5 < M < 15

test ordered hypotheses for multiple treatments, showed a significant decrease in individual differences from the group onset mean for close friend groups ($P \leq 0.001$). Second, a graph of this decrease as a function of time (Fig. 1) indicated that the greatest decrease occurred in the first 4 months with little subsequent change. This asymptotic relation indicated that the decrease in difference between onset dates was indeed an increase in synchrony for close friend groups.

Usually those who considered themselves close friends lived together. Because this was not always the case, however, subjects were divided into thirteen living groups ($5 \leq N \leq 12$), solely on the basis of arrangement of rooms, to test the importance of geographic location. When grouped in this way, there was no significant increase in synchrony within groups.

Dewan¹¹ has suggested that the menstrual cycles of monkeys around the equator are synchronized because each cycle is locked in phase with the Moon. As the production by the pineal gland of a substance which inhibits the action of luteinizing hormone is suppressed by light, the continuous light of nights with a full Moon would facilitate ovulation across a group of monkeys and induce synchrony. This suggests that the synchrony in close friend groups and among room mates comes from a common light-dark pattern, perhaps with common stress periods in which the subjects may stay up for a large part of the night. It would be expected that if synchrony arose from common light-dark cycles, room mates would exhibit a more significant amount of synchrony than do closest friends. The opposite trend was found, however, although it was not significant (room mates, $P \leq 0.007$; closest friends, $P \leq 0.003$). It does not seem likely therefore that a photo-periodic effect is a significant cause of synchrony. This is further supported by the lack of significant synchrony in random pairings in the dormitory.

Paralleling the Whitten effect in mice³ (in which suppression of oestrus in groups of females can be released by the introduction of a male pheromone) synchrony may result from a pheromonal interaction of suppression among close friend groups, followed by a periodic release due to the presence of males on the weekend. However, this would be insufficient to explain the synchrony which occurred among room mates and close friends, but did not occur throughout the dormitory.

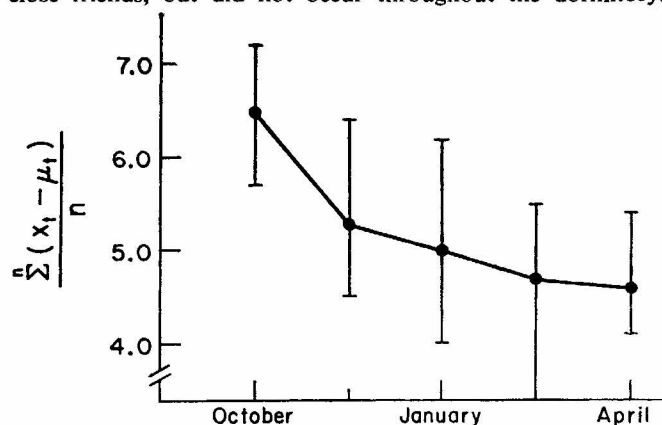


Fig. 1 The median individual mean difference from the group onset mean ($\frac{\sum (x_i - \mu_t)}{n}$ days) as a function of time. The asymptotic relation and non-overlapping confidence intervals⁸ for the medians in October and late February, and October and April (> 0.99), indicate an increase in synchrony for close friend groups.

Some additional pheromonal effect among individuals of the group of females would be necessary. Perhaps at least one female pheromone affects the timing of other female menstrual cycles.

Another possible source of synchrony might be the awareness of menstrual cycles among friends. A sample taken from the dormitory, however, indicated that 47% were not conscious

Table 2 Mean Cycle Lengths and Duration of Menstruation

Estimated exposure to male (days/week)	Length of cycle (days)	Duration (days)
0-2 N = 56	30.0 ± 3.9	5.0 ± 1.1
3-7 N = 31	28.5 ± 2.9	4.8 ± 1.2
P	≤ 0.03	N.S. ≤ 0.2

of their friends' menstrual cycles, and, of the 53% who were, 48% (25% of the total) were only vaguely aware.

The significant factor in synchrony, then, is that the individuals of the group spend time together. Whether the mechanism underlying this phenomenon is pheromonal, mediated by awareness or some other process is a question which still remains open for speculation and investigation.

Subjects were divided into two groups: those who estimated that they spent time with males, once, twice or no times per week ($N = 42$), and those who estimated that they spent time with males three or more times per week ($N = 33$). Borderline cases and those taking birth control pills were discarded. After testing for homogeneity of variance, the mean cycle length and duration of menstruation was compared using Student's t test. Those who estimated seeing males less than three times per week experienced significantly ($P \geq 0.03$) longer cycles than those of the other group whose mean cycle length corresponded with national norms (approximately 28 days)¹². There was no significant difference in duration of menstruation itself ($P \geq 0.2$, Table 2).

The possibility that the results were confounded by a maturational factor was tested, as subjects included members of the freshman, sophomore, junior and senior classes. The subjects were regrouped and compared according to class: underclassmen were compared with upperclassmen. There was no significant difference in cycle length (underclassmen 29.6 ± 5.6 days; upperclassmen 29.9 ± 5.7 days).

Exposure to males may not be the significant factor. It may be, for example, that those with longer cycles are less likely to spend time with males. However, many subjects spontaneously indicated that they became more regular and had shorter cycles when they dated more often. For example, one subject reported that she had a cycle length of 6 months until she began to see males more frequently. Her cycle length then shortened to 4.5 weeks. Then, when she stopped seeing males as often, her cycle lengthened again. Whether this is due to a pheromone mechanism similar to the Lee-Boot effect in mice¹ has yet to be determined.

Although this is a preliminary study, the evidence for synchrony and suppression of the menstrual cycle is quite strong, indicating that in humans there is some interpersonal physiological process which affects the menstrual cycle.

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LETTERS TO NATURE

PHYSICAL SCIENCES

Mechanism for Cosmic Ray Modulation

It is commonly believed that the basic mechanism responsible for the 11 yr solar cycle variation in cosmic ray intensity is well understood. We wish to show that on present evidence there is still a major puzzle concerning the dynamics of the process.

An approximate solution to the Fokker-Planck equation which expresses the balance between cosmic ray diffusion into the solar system and removal by a combination of convective sweeping by the solar wind and energy loss of particles in an expanding medium is:

$$\frac{n_e}{n_s} = \exp - \left[\frac{V r_s^3 \left(1 + \frac{\gamma - 1}{3} \right)}{3 D_{\parallel} r_e^2} \right] \quad (1)$$

where n_e and n_s are respectively the differential cosmic ray density in a magnetic rigidity range $P \rightarrow P + dP$, at the Earth's orbit (1 AU) and outside the effective boundary, r_s , to the modulating region. Diffusion, with a coefficient $D_{\parallel}$, is confined to the Archimedian spiral field direction and this direction is close to azimuthal in the ecliptic plane. Further assumptions are that $r_s \gg r_e$, $D_{\parallel}$ is proportional to magnetic rigidity P , but independent of radial distance for the greater than 1 GV rigidity particles under discussion and $n_s \propto P^{-\gamma}$. V is solar wind velocity. Jokipii¹ has suggested that in the 1 to 10 GV rigidity range $D_{\parallel}$ is directly proportional to the total power spectral density in the fluctuations of the interplanetary magnetic field components at a wavelength in resonance with the helical motion of the particles about the mean Archimedian spiral field.

From equation (1), it would be expected that over the solar activity cycle, a direct correlation should exist between low

energy cosmic ray intensity and at least one of the three parameters of V , $D_{\parallel}$ and r_s which specify the depth of modulation. Data obtained over the past several years, however, suggest that this is not the case. Fig. 1 shows the Deep River neutron monitor intensity variation from 1962 to 1969. The monitor responds to cosmic rays above 1 GV rigidity, but with a maximum response at ~ 6 GV. The changing level of solar activity of this period is shown by means of smoothed Zurich sunspot numbers. Also shown in the figure are various solar wind speed measurements carried out by Mariner-2², IMP-1³, VELA-2⁴, Pioneer-6 and 7⁵, and HEOS-1⁶ satellites. The solar wind speeds show no systematic long term variations.

The apparent lack of correlation between V and cosmic ray intensity is shown more explicitly in Fig. 2 in which modulation index $\eta(t)$ is plotted against V . $\eta(t)$ is defined by the simplified form of equation (1):

$$\frac{n_e}{n_s} = \exp - \left[\frac{\eta(t) + \eta(t_0)}{P} \right] \quad (2)$$

with P in GV. $\eta(t_0)$ is the value of the modulation index at solar minimum. Previous experimental studies have established the rigidity dependence of the exponent and $\eta(t_0)$ is generally taken to be less than 1 GV. $\eta(t)$ used in Fig. 2 were derived from percentage changes of the Deep River neutron monitor intensity from the solar minimum value, using differential response sanctions of sea level neutron monitors^{7,8}.

Also in Fig. 2 is a plot of power spectral levels totalled over the three components of the interplanetary magnetic field and measured at 1 a.u. during 2 months by Mariner-2⁹, during 1 month by Mariner-4¹⁰ and during 3 months by the Imperial College magnetometer on HEOS-1. (Published data from Pioneer-6 have been neglected for reasons given by Belcher and Davis¹¹.) The 1 GV cosmic rays suffer resonance scattering in a mean quiet-time field of 3.6γ at 1.3×10^{-4} Hz. Fig. 2 shows the power levels at two convenient frequencies, that is 4.6×10^{-5} Hz and 1.7×10^{-4} Hz. If $D_{\parallel} \propto P f(t)$, variations in the

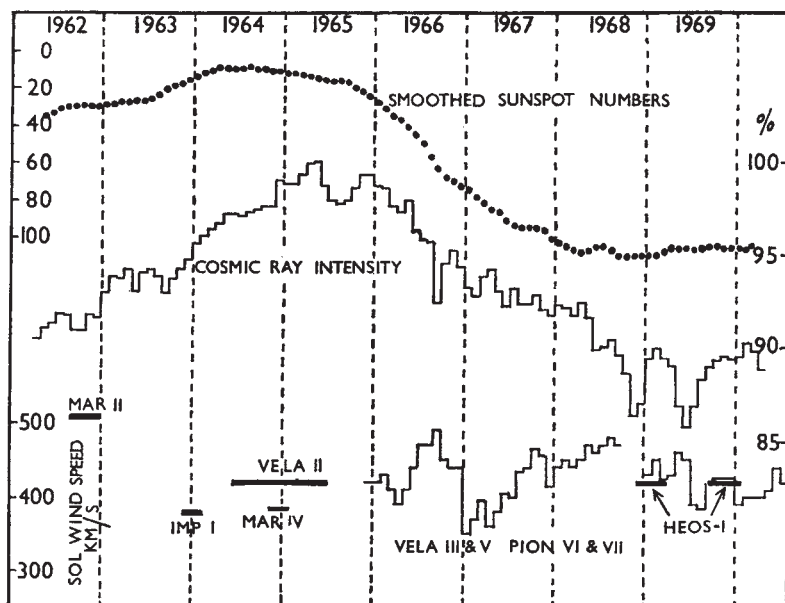
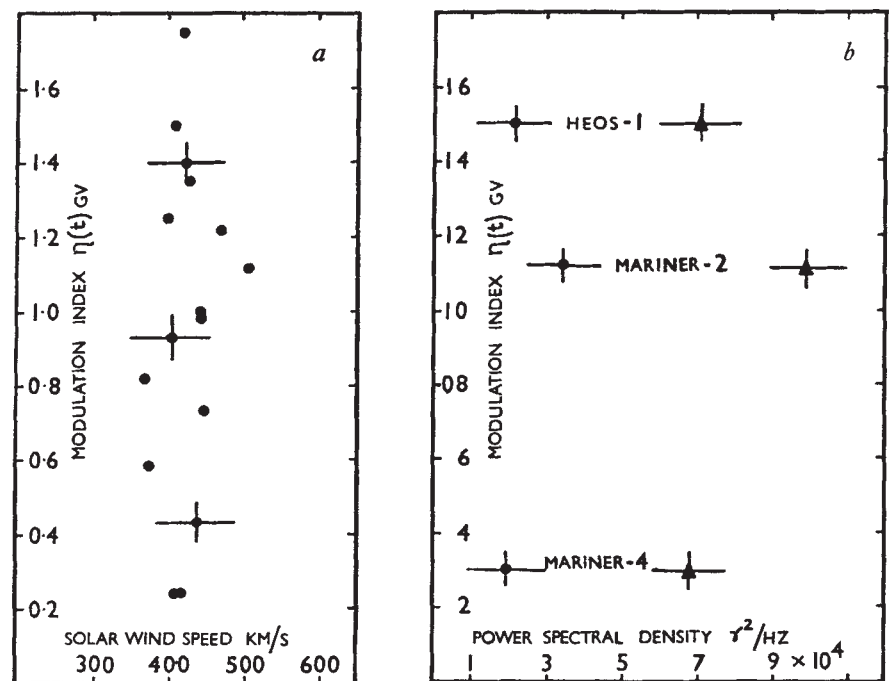


Fig. 1 Smoothed sunspot numbers, Deep River neutron monitor data normalized to May 1965 counting rates and solar wind data, from 1962 to 1969.

Fig. 2 *a*, Cosmic ray modulation index versus average solar wind speed (averaged over four month periods). *b*, Cosmic ray modulation index versus power spectral density of interplanetary magnetic field fluctuations at the frequencies of 4.6×10^{-5} Hz ($\blacktriangle$) and 1.7×10^{-4} Hz ($\bullet$).



power levels at these frequencies should be representatives of variations in $D_{||}$ throughout the 1–10 GV rigidity range. Fig. 2 shows that the power spectra do not change in a manner that is required to account for the changes in cosmic ray intensity.

The effective boundary of modulation region r_s , the third relevant variable parameter, is unlikely to change sufficiently to account for the cosmic ray modulation. If this boundary is taken to be where the solar wind is stopped by the galactic magnetic field, that is at about 50 a.u., it is unlikely to change much as neither solar wind speed nor solar wind density⁶ does as required, making solar wind energy density more or less invariant over a long period. Furthermore, if the boundary is as distant as 50 a.u., it would suggest a very large residual modulation at solar minimum and consequently a very high value to the galactic cosmic ray intensity¹². The boundary is more likely to be at the position where interplanetary field waves grow to the same size as the mean field value and create a turbulent shell which is relatively ineffective in causing modulation because the mean spiral field configuration is gone. Belcher, Davis and Smith¹³ find that much of the wave structure in the interplanetary field is due to propagation in the Alfvén mode where the relative wave growth is given by

$$\frac{\nabla B}{B} = \left(\frac{\nabla B_e}{B_e} \right) \frac{r}{r_e} \quad (3)$$

We have already shown representative power levels of fluctuations which remain rather steady over several years. The mean field modulus does not seem to change either; for example, $\langle |B| \rangle$ is $6.3 \pm 0.3 \gamma$ from HEOS-1 data compared with an IMP-1 value of $6.0 \pm 0.25 \gamma$ ¹⁴. Thus any boundary defined by relative wave growth is unlikely to be very solar cycle dependent.

If then the cosmic ray modulation over the cycle of solar activity cannot be explained by observed variations in solar wind and interplanetary magnetic field parameters, what alternatives can be offered? Possibilities include the following. (1) The residual modulation index $\eta(t_0)$ is much greater than unity, hence allowing only small changes in V or spectral power to bring about the necessary variation in cosmic ray intensity. If this were true, current ideas on the cosmic ray energy density, including electrons in interstellar space, would need revision. (2) The solar cycle variation is a summation of Forbush decrease events caused by rather ordered field configurations which do not show up in the power spectral levels.

The possible contributions from Forbush decreases in an old idea¹⁵, unresolved at present. (3) Off-ecliptic phenomena may be important. For example, changes in relevant parameters at the heliographic latitude opposite the zone of maximum sunspot activity may be chiefly instrumental in determining the effective position of the boundary of the modulation region. Verification of this requires measurements by off-ecliptic space probes. Present difficulties in explaining the dynamics of the 1 yr cosmic ray modulation support arguments for the addition of such probes to the present programme of exploration in the distant solar system.

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Goethe on Interferometry

I HAVE had occasion recently to re-read Goethe's *Theory of Colours* (1810) and have found that he described the nature of the microtopographical interference pattern given by a crystal surface, presumably from the description, that of a cleavage. In chapter 33, which deals with interference colours (Goethe calls these epoptical colours), first a precise account is given of the (already known) appearances of white light, Newton's rings produced when glass lenses are pressed together. Colour sequences at various pressures are described in detail.

Goethe then reports that the pressing of plane [*sic*] glasses together produces colours which "undulate like watered silks", a perfect description of the typical wavy form of interferogram given by polished glass plates (Fig. 1). These colours he traces through under varying pressures and varying angles of incidence. He notes that clear fringes only arise with clean glass, which, he advises, should be handled with gloves, and even recommends observation in a vacuum. The effects, he rightly states, are most brilliant when concave and convex surfaces of the same curvature are brought into juxtaposition, and thus his fringes are seen to advantage with achromatic telescope objectives. Goethe discusses in some detail the central reflected black spot and makes the acute observation that this reflected black spot is a very bright spot in transmission. Since he was not aware of wave theory and phase changes he makes the reasonable suggestion that the reflectivity at the pressure point has dropped to zero.

In paragraph 446 Goethe gives a remarkable description of the interference fringe pattern he sees with a crystal. He says, "A remarkable appearance takes place when dissimilar surfaces are pressed together, for example, a polished crystal and a plate of glass. The appearance does not exhibit large flowing waves, as in the combination of glass with glass, but is small and angular, and, as it were, disjointed".

In another section of the same treatise Goethe mentions that he has available a rhomb of Iceland spar. I venture to conjecture that this is the polished [*sic*] crystal he is using. There is not the slightest doubt that what he is describing is just what is seen when a well cleaved calcite crystal is matched against a piece of polished glass. There is just the alternative (but less likely) possibility that a natural crystal such as quartz with good growth features was being matched. However, the description of interference fringes as "small, angular and disjointed" is exactly what is revealed by a good cleavage surface like that of calcite (Fig. 2).

This really notable observation by Goethe seems to have been completely missed by both physicists and historians of the nineteenth and twentieth centuries. Could it be that this was due to the cold disdain by which Goethe's critique of Newton was received by contemporary and later scientists? Because the major colour theory was condemned, maybe the associated notable excellent observations were ignored.

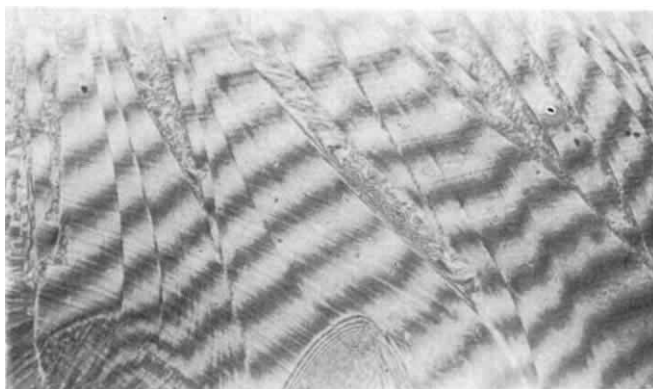


Fig. 2.

In succeeding sections Goethe gives notable descriptions of his penetrating observations on a very wide range of thin film interference fringes. All these effects he regretfully attempts to account for by his now discredited theory of the origin of colours. Nevertheless, theory apart, this fascinating text teems with novel interferometric observations, making the text a joy to read.

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X-ray Observation of Cen XR-4 and Nor XR-2

WE wish to present the results of an X-ray astronomy experiment conducted from a Centaure rocket launched from Thumba Equatorial Rocket Launching Station (TERLS), India, at 0035 UT on December 7, 1969. The detector consisted of a xenon-methane proportional counter of useful area of 55 cm² with a 18 mg cm⁻² thick beryllium window and a slat collimator with a field of view of 7° × 15°. It had a resolution of 20% for 6 keV X-rays from a ⁵⁵Fe radioactive source. The rocket aspect¹ determined from onboard magnetic and Sun sensors showed that it had a spin rate of 5.5 r.p.s. and a precession with a half cone angle of about 3° around an axis centred at R.A. 8 h 56 m ± 4 m and declination 3° ± 1°. The X-ray data were pulse height analysed into three contiguous energy channels in the energy range 2–18 keV.

Fig. 1 shows the plot of count rate in the 2–9 keV range as a function of spin azimuth. Various sources within the scan region of the detector are marked in the figure, the source positions having been obtained from earlier observations^{2–4}. As the figure shows, the fluxes from Cen XR-1, Cen XR-2 and Cen XR-4 are much below the limit of detectability and hence only upper limits of flux for these sources are derived (Table 1).

All available observations of Cen XR-4 at different epochs^{3,5,6} are plotted in Fig. 2. Data from Vela satellites used in this diagram have been corrected for transmission efficiency of the

Table 1 Upper Limits for the X-ray Flux from Cen XR-1, Cen XR-2 and Cen XR-4 in the Energy Range 2–18 keV (2 σ level)

Energy range (keV)	Flux in photons cm ⁻² s ⁻¹ keV ⁻¹		
	Cen XR-1	Cen XR-2	Cen XR-4
2–5	1.4×10^{-2}	3.6×10^{-2}	1.5×10^{-2}
5–9	0.95×10^{-2}	2.4×10^{-2}	1.0×10^{-2}
9–18	0.52×10^{-2}	1.3×10^{-2}	0.55×10^{-2}

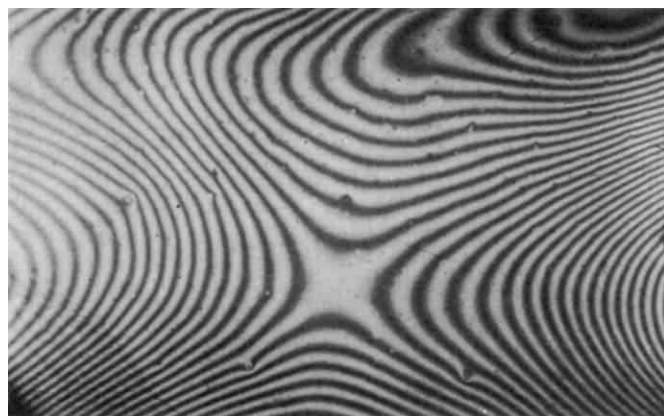


Fig. 1

detector window. Cen XR-4, which erupted suddenly as an X-ray star in July 1969, decayed to less than 0.5% of its peak intensity by September 24, 1969 (ref. 6). Our observation on December 7, 1969, yields an upper limit of $0.13 \text{ photons cm}^{-2} \text{ s}^{-1}$ for the flux from this source in the energy range 2–18 keV.

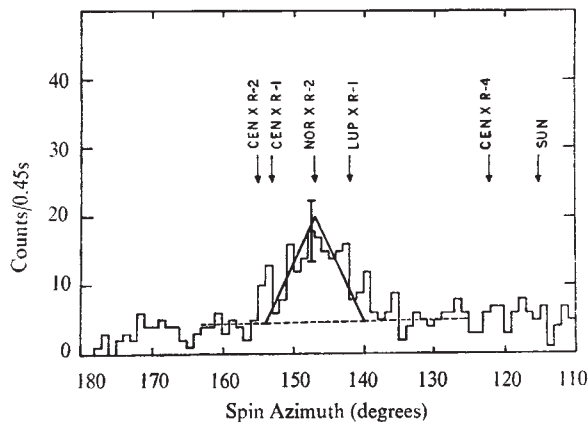


Fig. 1 Total counts in 2–9 keV range for 0.45 s plotted as a function of spin azimuth.

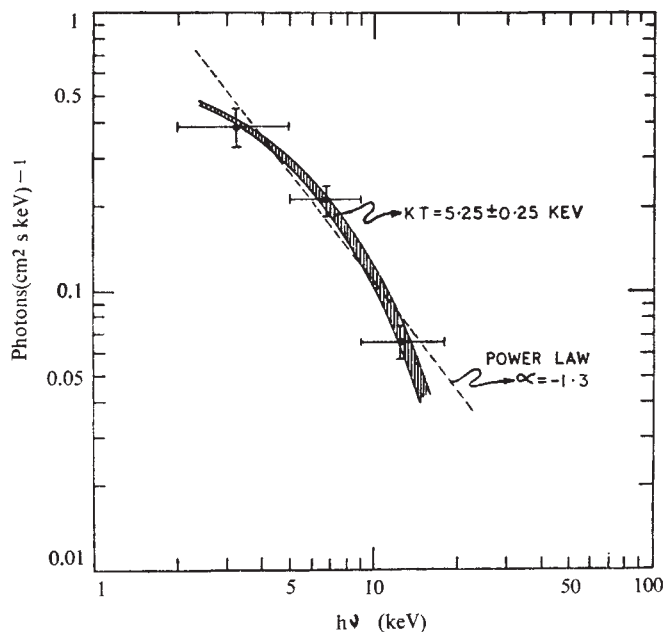


Fig. 3 The differential photon spectrum of the Nor XR-2 source.

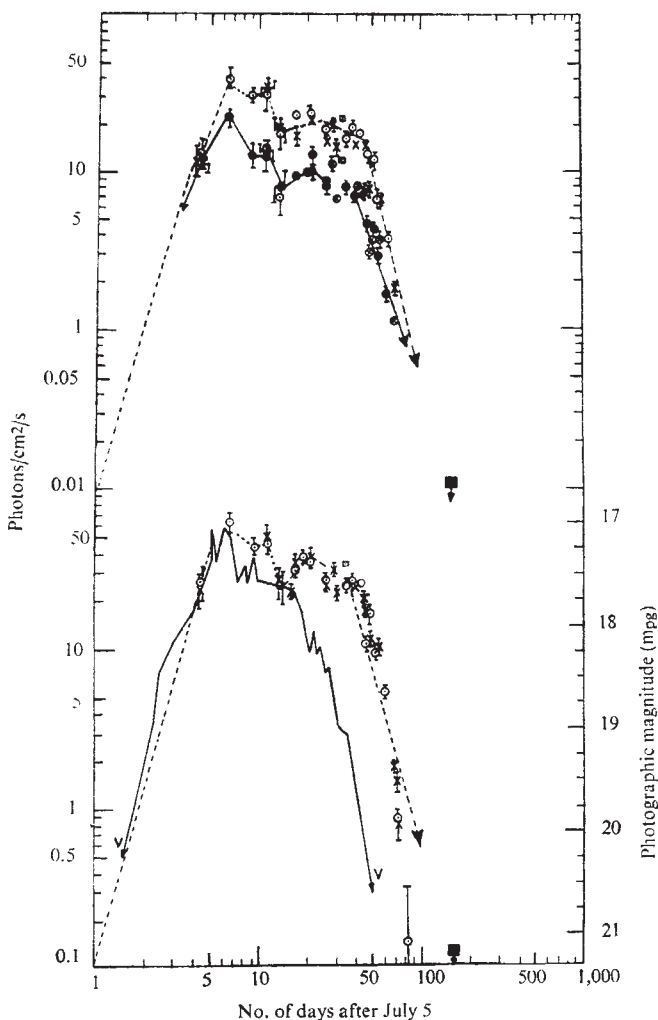


Fig. 2 X-ray flux from Cen XR-4 as a function of time after July 5. The lower curves show the light curve of nova No. 13 in Andromeda nebula (—) and the X-ray flux in the energy range 3–12 keV (---). The upper curves show X-ray flux from the same source in two energy bands 3–6 (---) and 6–12 (—) keV. ⊗, ●, ○, ×, Vela (Evans *et al.*)⁶; □, Kitamura *et al.*⁵; ■, present experiment.

The similarity between the concentration of X-ray sources and that of novae along the galactic plane has already been pointed out by L. Gratton (at the IAU symposium on non-solar γ and X-ray astronomy in Rome in 1969). The spectacular time variation, shown by X-ray sources like Cen XR-2 and Cen XR-4⁶, is very much like the optical luminosity variation of novae. Arp⁷ has established a definitive relationship between the maximum magnitude attained by novae and their total duration, the lesser magnitude novae corresponding to lesser duration. Novae with a duration similar to Cen XR-4 will have a typical apparent magnitude of about 17. Since the novae in the galaxy M31 have the same characteristics as our galactic novae, we have plotted in Fig. 2 the light curves for No. 13 nova (as L. E. Peterson said, also at the Rome symposium in 1969) in M31 which has an apparent magnitude of about 17. The similarity between the light intensity of a typical fast nova and the X-ray flux of Cen XR-4 is very striking, indicating that Cen XR-4 is a nova-like object.

Considering the triangular response of the collimator, the main peak in the count rate (Fig. 1) can be attributed entirely to Nor XR-2 at R.A. $234^\circ 30' \pm 2'$ and declination $-56^\circ 22' \pm 2'$ consistent with the position coordinates given by Friedman *et al.*².

Even though Chodil *et al.* observed a flux of $\sim 1.6 \text{ photons cm}^{-2} \text{ s}^{-1}$ (2–10 keV) for Lup XR-1 in 1967, subsequent low energy observations by Peterson (reported in Rome) and MacGregor *et al.*⁸ have failed to confirm this. Further, the high energy balloon observation of Lewin *et al.*⁹ has indicated the absence of this source. In spite of the reported location of Lup XR-1 only 3° away from collimator normal as compared with 8° for Nor XR-2, we do not observe a significant flux (at 2σ level) from the direction of Lup XR-1, which taken with Chodil *et al.*'s⁴ observation indicates that Lup XR-1 is also a time varying X-ray source.

The counts in different energy channels 2–5, 5–9, 9–18 keV for Nor XR-2 source were obtained by subtracting the background, which was derived from the mean counts on both sides of the source position. The absolute flux $N_p(E)$ was derived from the count rates by folding the efficiency $\epsilon(E)$ of the detector and the atmospheric transmission along the line of sight $Tr(E)$ as

$$N(E_1, E_2) = \int_{E_1}^{E_2} N_p(E) Tr(E) \epsilon(E) dE$$

and considering the geometrical response of the detector system. In the energy range 2–10 keV, we observe a flux of 2.0 ± 0.3 photons $\text{cm}^{-2} \text{s}^{-1}$ for the Nor XR-2 source, which can be compared with the value of ~ 2.6 photons $\text{cm}^{-2} \text{s}^{-1}$ obtained by MacGregor *et al.*

Fig. 3 shows the plot of the differential photon flux for Nor XR-2 which can be fitted to either an exponential spectrum $N_p(E) = 0.75 e^{-E/5.25 \pm 0.25}$ photons $\text{cm}^{-2} \text{s}^{-1} \text{keV}^{-1}$ corresponding to a hot thin plasma at 6×10^7 K or to a power law spectrum $N_p(E) = 2.1 E^{-1.3 \pm 0.2}$ photons $\text{cm}^{-2} \text{s}^{-1} \text{keV}^{-1}$.

Even though a power law spectrum is generally considered as indicative of the synchrotron mechanism for X-ray production, Manley¹⁰ has shown that consideration of a sharp high energy cut off in the relativistic electron spectrum can modify the power law spectrum for X-rays into an exponential one. It is extremely interesting to note that a supernova remnant¹¹ p 1548–55 at R.A. $237^\circ 15'$ and decln $-55^\circ 59'$ and a pulsar¹² MP 1530–53 at R.A. $232^\circ 35' 45'' \pm 30''$ and decln $-53^\circ \pm 1^\circ$ are located within the uncertainty circle of Nor XR-2 coordinates. Since Poveda and Woltjer¹¹ first suggested the possibility that all supernova remnants were X-ray emitters, only two X-ray sources have been definitely identified with supernova remnants. The likelihood of Nor XR-2 being also associated with a supernova remnant, coupled with the possibility of its X-ray spectrum favouring a synchrotron emission, makes this a source of great astrophysical interest.

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Deposition of Molybdenum and Uranium along the Major Ocean Ridge Systems

ANOMALOUSLY high concentrations of uranium and molybdenum along with other transition metals have been found in the surface sediments on the world-wide system of rises^{1–5}. Bostrom *et al.*^{1,2} ascribe the high transition metal content of the East Pacific Rise sediments in particular to volcanic activity. We suggest here an alternative hypothesis, at least for uranium and molybdenum and possibly for some of the other elements.

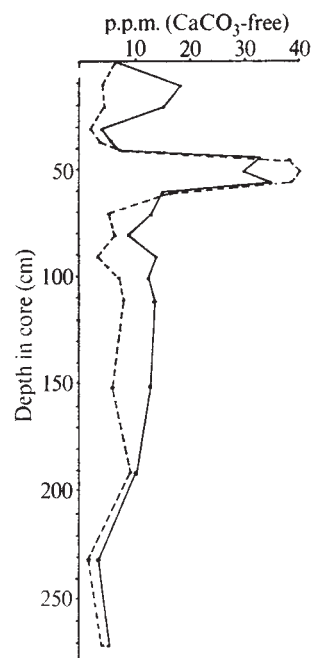


Fig. 1 The variation of uranium (---) and molybdenum (—) down the length of Lamont-Doherty core A180-74 ($00^\circ 03' \text{ S}$, $24^\circ 10' \text{ W}$) on a CaCO_3 -free basis.

Molybdenum has been shown to be enriched in the surface sediments of the mid-ocean seismic ridges and also the secondary relatively aseismic ridges such as the Rio Grande Rise and Walvis Ridge.⁴ The work of Bostrom and Peterson¹ on the East Pacific Rise indicates that the concentration of molybdenum is not peculiarly high along the areas of high heat flow. This implies that a volcanic origin may not be the best explanation. The pattern of uranium distribution along the world's ridge systems is less well known but some information bearing on the source of uranium is available from isotopic results. The $^{234}\text{U}/^{238}\text{U}$ activity ratio of the top layer of a high-uranium concentration core on the East Pacific is 1.16, suggesting that the uranium was derived from seawater⁵.

In addition to the regional variation patterns the variation with time at a location is useful in determining the source of the metals. In order to investigate the time varying pattern of the deposition of uranium and molybdenum along a ridge site we have studied^{3,4} the distribution of these elements with depth in a core raised from the Mid-Atlantic ridge—Lamont-Doherty core A180-74. This core had previously been examined for Mn, Fe, Cr, Pb, Sn, Ni, Co, Sb, and Ag (refs. 6 and 7, and unpublished work of K. K. Turekian and D. P. Kharkar). The results for uranium and molybdenum are presented in Fig. 1. The two elements vary sympathetically with a pronounced enrichment of uranium and molybdenum at the depth interval between 40 and 70 cm. Above this interval the core is brown in colour with high concentrations of manganese suggestive of oxidizing conditions but without any marked enrichment of uranium and molybdenum. In the interval between 35 and 70 cm the sediment was described in the shipboard log as dark grey in colour which turned lighter with age. Manganese is generally depleted in this interval. Neither iron nor chromium concentrations show any large variations down the length of the core.

The very high concentrations of uranium and molybdenum, accompanied by slight enrichments in silver, antimony and nickel in the 40–70 cm interval relative to adjacent intervals of core A180-74, indicate a common mechanism of deposition for these elements. This assemblage is characteristic of the enriched elements found in near-shore anaerobic sediments whose higher trace metal contents clearly derived from sea-

water^{8,9}. It seems likely therefore that the sympathetic enrichment of molybdenum and uranium in this deep sea core is also the result of deposition from seawater under anaerobic conditions in the sediments or at the sediment-water interface.

We propose that most of the sediments with high concentrations of molybdenum and uranium found on seismically active and relatively inactive ocean rises reflect the formation of anaerobic sediments, in part the result of the formation of local ephemeral basins along the ridges and in part the result of the high rate of supply of organic material to the relatively shallow ocean bottoms along the ridge area.

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Shock-tube Study of the Nucleation of Lead Vapour

THE homogeneous nucleation of supersaturated vapours is at present receiving much theoretical attention (see, for example, ref. 1). Experimentally, the phenomenon is difficult to examine in controlled and well defined conditions. One important experimental problem is the interference that can arise from dust and ions which act as rival nucleating sites, but this can be avoided by studying highly supersaturated vapours in which the rate of homogeneous nucleation is rapid. In the past this approach has been limited to research on expansion nozzles. Jaeger *et al.*² have described such experiments and have reviewed the general situation.

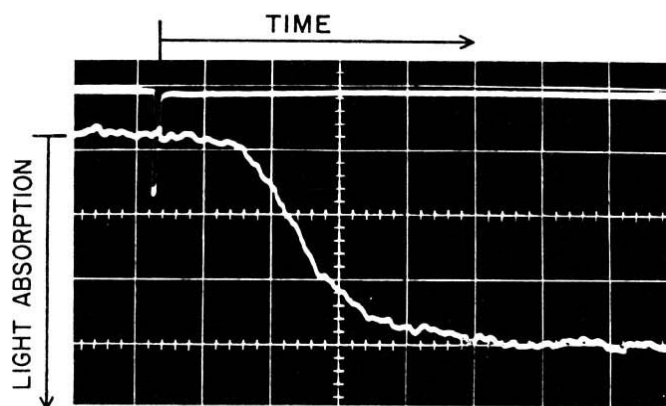


Fig. 1 An oscilloscope record showing, in the lower trace, the absorption of light at 433 nm by precipitating lead. The incident light intensity would be equivalent to 15 large divisions of the graticule. Each large division in the horizontal direction represents 140 μ s of gas time and the upper trace indicates the time of arrival of the shock wave at the observation window. Gas conditions are 935 K and $[TML]_{t=0} = 7.0 \times 10^{15}$ and $[Ar] = 1.4 \times 10^{19}$ molecule cm^{-3} .

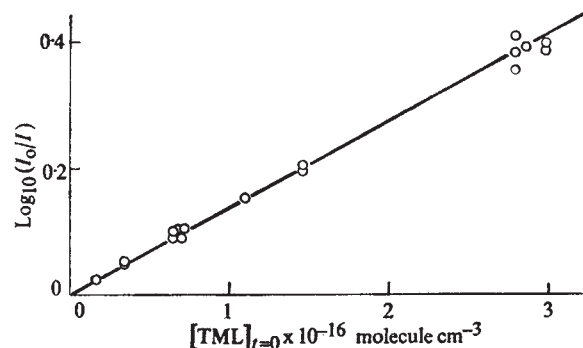


Fig. 2 The linear dependence of the light absorption, in the plateau region of the signal from the lead particles, on the total lead concentration in the gas.

During studies related to the antiknock action of lead alkyls in gasoline engines, we have been able to record precisely the rate of precipitation of lead from its highly supersaturated vapour. The technique used ensures that the gas is held at an essentially constant temperature during the precipitation process, so that it has a distinct advantage over the expansion nozzle technique and seems to be most suitable for a detailed study of nucleation phenomena.

The work was performed on a shock tube which has been described previously^{3,4}. Argon containing 125 to 2,000 p.p.m. tetramethyl-lead (TML) was shock-heated to temperatures of 600–1,200 K and pressures of 100–400 kN m^{-2} (1 to 4 atmospheres). The reactant material was sufficiently dilute for the temperature to be effectively constant over the experimentally available reaction time of about 5 ms. The TML pyrolysed directly to lead vapour which could become supersaturated and subsequently precipitate.

Flash absorption spectroscopy⁴ on the reactant gas at 900 K confirmed that considerable quantities of lead atoms were generated early in the reaction, and that later particles of lead were formed which scattered and absorbed the incident light beam characteristically more in the ultraviolet than in the red region of the spectrum.

The rate of precipitation of lead was recorded in subsequent experiments by monitoring the obscuration of light at a wavelength of 433 nm. An oscilloscope trace (Fig. 1) shows an induction period during which TML decomposed and the nucleation of lead vapour was initiated. This was followed by a rapid decrease in transmission, indicating the growth of precipitated lead, and finally an absorption plateau which continued for the remainder of the 5 ms of available time. The trace in Fig. 1 was obtained for a gas temperature of 935 K. At 1,000 K the process was even faster, the plateau being reached within 0.3 ms. For the same initial concentration of TML, however, a further increase in temperature resulted in a dramatic change in the signal record to one of much decreased strength and growth rate, indicating that the concentration of lead in the gas was insufficient, relative to its saturated vapour pressure at that temperature, to precipitate quickly. The temperature of the transition defined in broad terms the level of supersaturation necessary to promote rapid nucleation. Over the experimental range of initial concentrations of TML of $(0.17 \text{ to } 2.8) \times 10^{16}$ molecule cm^{-3} , the transition was found to occur consistently at a lead supersaturation ratio of 50 ± 10 .

As shown in Fig. 1, the growth of the condensed phase can be very rapid, in which case the lead particles will be very small and at high number densities. They will tend to coalesce on the time-scale of these experiments. The fact that the absorption signal becomes constant with time means that it is insensitive to this coalescence process and must therefore be a measure simply of the mass of condensed lead and not of the particle size. This was confirmed by the linear relationship

between the absorption in the plateau region (expressed as $\log(I_0/I)$) and the initial concentration of TML (Fig. 2), and is theoretically acceptable within the context of Mie theory of light scattering for a restricted range of wavelength⁵.

The rate of decomposition of TML has been measured (J. B. H. and I. R. H., unpublished), so that the rate of release of lead atoms to the gas phase could be calculated and applied, together with simple kinetic theory of particle growth, to the rate of condensation observed. Such a calculation showed that the diameter of the particles at the start of the plateau (Fig. 1) was less than 4 nm, with a corresponding particle density greater than $5 \times 10^{12} \text{ cm}^{-3}$ and a rate of nucleation peaking at $2 \times 10^{16} \text{ cm}^{-3} \text{ s}^{-1}$.

The research has demonstrated the potentialities of this technique and is being pursued.

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Behaviour of Single Wood Fibres under Axial Tensile Strain

INTEREST has arisen recently in the stress-strain properties of single wood fibres and tracheids, because of their importance to the properties of paper, board and other composite structures. Techniques for handling and mounting these fibres, which are only about 1 mm in length and 30 μ in diameter, have been developed in several laboratories and apparatus for

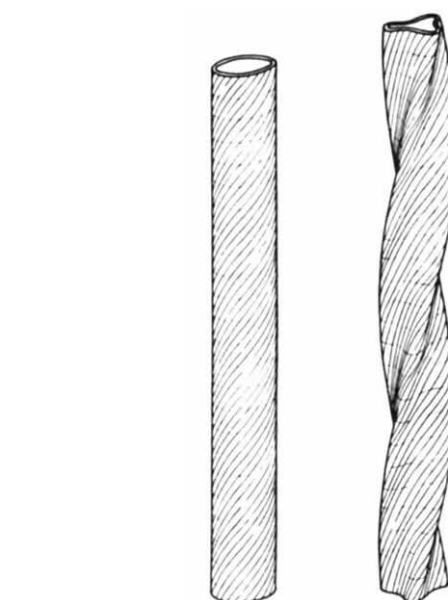


Fig. 3 Left, spirally wound tube which, according to theory, buckles under axial tensile strain to the form indicated on right.

determining stress-strain curves has been built. A survey of recent work is given by Duncker and Nordman¹. To provide for simultaneous microscopical observation of the strained fibre, an apparatus was built incorporating an Instron tensile tester and a Zeiss research microscope (K. W., R. Bain and D. H. P., unpublished work). The preliminary results of this study are reported here and indicate that the deformation of wood fibres under uniform axial strain is by no means a simple matter.

The phenomenon illustrated in Fig. 1, which shows a fibre before and after straining, was observed in many fibres. The fibre appears to have twisted under strain, even though its ends have remained firmly glued to their non-rotatable supports. A similar but more striking example is shown in the scanning electron micrograph of a strained fibre in Fig. 2. The develop-

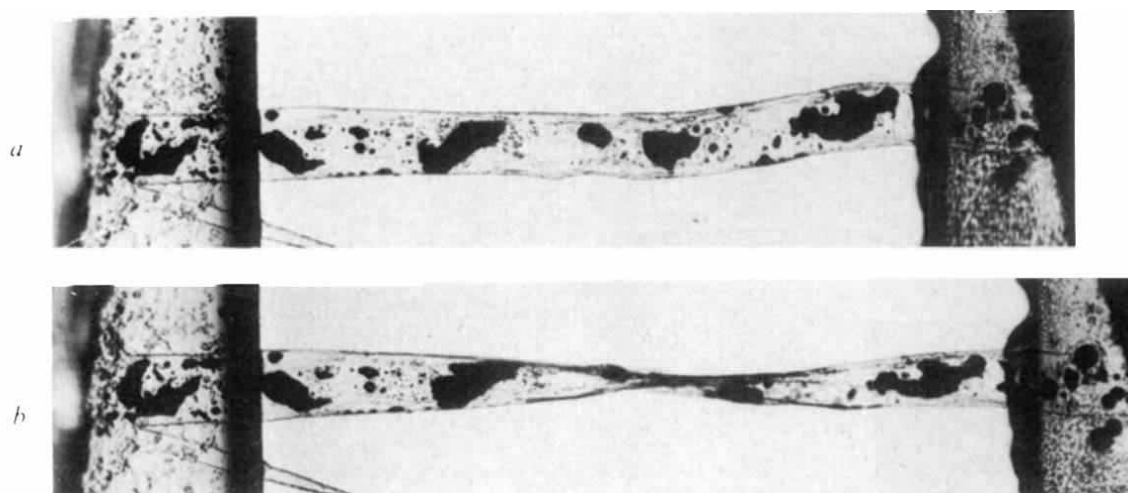


Fig. 1 Sprucewood fibre, pulped by the kraft process. *a*, Unstrained; *b*, strained. The strained fibre appears twisted even though it is clear that the ends have not rotated. The dark areas are mercury droplets introduced into the fibre lumen to permit fibril angle measurement². ($\times 145$.)

Fig. 2 Scanning electron micrograph showing apparent twists in strained fibre. ($\times 200$.)



ment of this twisted appearance has been observed many times under the microscope and recorded on ciné film.

This can be explained in terms of the structure of wood fibres and the theory of buckling of orthotropic shells. Wood fibres are hollow tubes composed of layers of cellulosic protofibrils embedded in a matrix of hemicellulose and lignin. Most of the cell wall material is contained in the S_2 layer, the protofibrils of which trace a steep spiral around the fibre. The wood fibre can thus be considered as a spirally wound fibre reinforced composite tube. Detailed studies of the properties of such tubes have been carried out by materials scientists in the aerospace industry³. In particular Pagano, Halpin and Whitney⁴ have considered their stability under axial tensile strain and have shown that, because of induced shear stresses, buckling can occur. They showed both experimentally and theoretically that the mode of buckling is as illustrated in Fig. 3. The similarity of this form to the one observed in single wood fibres is apparent.

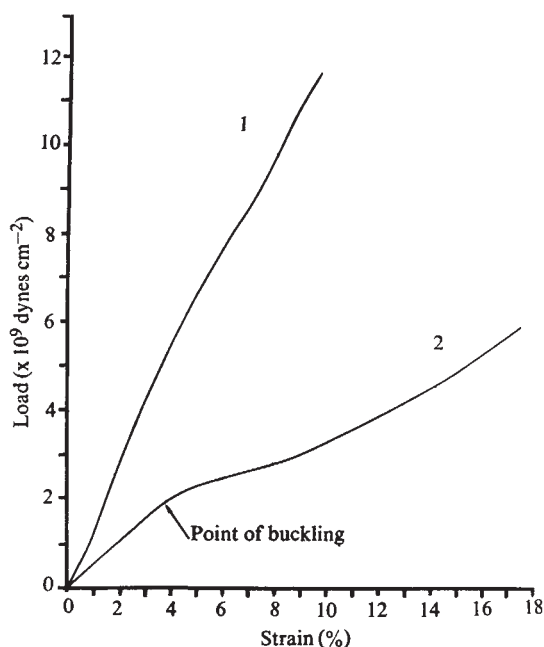


Fig. 4 Stress-strain curves of sprucewood fibres. 1, A fibre that does not buckle; 2, a fibre that buckles at the point indicated.

Analysis of the ciné films and stress-strain records shows that the shape of the stress-strain curve is influenced by the onset of buckling. Fibres that exhibited no observable buckling had substantially linear stress-strain relations, but fibres that buckled gave sigmoidal curves of the form shown in Fig. 4. The first appearance of the buckled shape coincided with the yield point of the stress-strain curve. Buckling theory predicts that the critical buckling stress is a function of the principal elastic constants of the fibre wall, the angle of the spiral winding and the fibre wall thickness. Preliminary data indicate that the critical stress can be predicted in this way and thus it seems that a quantitative application of buckling theory will give a more precise description of the stress-strain curve. This approach is being pursued.

The implications of these findings are technologically important. The stress-strain curves of fibres that are prevented from buckling are quite different from those of free fibres. Thus, while data obtained from isolated fibres are relevant to the behaviour of loosely bonded structures such as non-woven fabrics, they are not directly applicable to the behaviour of tightly bonded structures such as paper and board. The phenomenon may also have general interest. Spirally wound structures commonly occur in nature, for example, in seed

hairs such as cotton, and these may provide other examples of this type of deformation.

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BIOLOGICAL SCIENCES

Was Virchow Right about Neandertal?

IVANHOE'S¹ claim that "Virchow diagnosed rickets in the Neandertal bones, accounting so for their peculiar simian cast," is rather misleading. All Virchow² stated was that the skull was that of an individual suffering from rickets and osteoarthritis and therefore racially unclassifiable. Since the antiquity of the Neandertal fossil is now no longer in doubt, the statements of Virchow and Ivanhoe raise two separate questions.

First, did Neandertal man (as a race) suffer from vitamin D deficiency? This has been argued for several fossils on the basis of evidence that has been summarized by Ivanhoe¹.

The next question is: are the diagnostic characters of the Neandertal race due to rickets? Straus³ has examined the La Chapelle skeleton and shown that some of the characters noted by Boule as typical of Neandertal are due to chronic osteoarthritis. But the total morphological pattern of the skull and post-cranial skeleton, which is so strikingly characteristic of this European race, is widely distributed in time and place and clearly occurs in the lower latitudes where vitamin D deficiency would not be expected. Furthermore, the skulls of the Riss-Würm interglacial, when the climate was presumably even more pleasant than today, have the Neandertal characters sufficiently pronounced so that their taxonomic assignment and evolutionary status are not in doubt. This forces Ivanhoe to classify Ehringsdorf, Fontéchevade, and Saccopastore as *Homo erectus* which is justified neither by their morphology nor by their age. It is now recognized that the species *Homo sapiens* is of at least Riss age (to include Swanscombe and Steinheim), and Ivanhoe can avoid consideration of the steady evolution of Neandertal morphology in Europe during the warm Riss-Würm period only by the taxonomic scheme proposed by him.

Ivanhoe does not seem to appreciate fully the nature of geographical variation. Factors other than latitude and altitude are always at work, even though we may not recognize them. Geographical variation is so extensive at this stage of human evolution that whether they are contemporary or not it is necessary to separate taxonomically the Chinese (Mapa), south-east Asian (Solo), the South African fossils (Broken Hill and Saldanha) and the European Neandertalers. Clines certainly existed throughout these areas but can only be properly recognized among contemporary forms. It is equally dangerous to postulate evolutionary lineages from Krapina to La Ferrassie, or from Solo to Niah. What we have for study are very small samples from local populations, the origin and fate of which are not known.

Ivanhoe argues that the incidence of rickets fell away with the passing of the cold phase of Würm I. The archaeological

evidence suggests, however, that Neandertal was replaced by modern man at the peak of the cold phase. Furthermore, we know that modern forms survived through succeeding cold stadia without regaining "Neandertal" morphology, in areas where the exploitation of fish as a food source was unlikely to be a significant source of vitamin D.

Ivanhoe states that "Virchow's original hypothesis can be tested empirically". We claim that it cannot, by the method proposed by Ivanhoe. All the suggested tests can do is to determine the presence or absence of, as well as the severity of damage due to, vitamin D deficiency. This in no way proves that the taxonomic characters of Neandertal man are due to such deficiency. For example, Ivanhoe states that one of the diagnostic characters of rickets in children is "a high bulbous forehead (olympian front)" while the most characteristic feature of Neandertal man is precisely the opposite: a fleeting forehead with the total absence of a bulbous frontal. In so far as the Neandertal child has a more bulbous frontal than the adult, this can be satisfactorily explained from our knowledge of skull ontogeny in man and other higher primates.

Neither does the parallelism between the appearance of Neandertal characters and the Arctic climate prove that the characters are caused by vitamin D deficiency. If the characters of Neandertal man are of adaptive significance, and with Clark Howell⁴ we agree that this is so, at least in part, then we can expect them to be correlated with climate and geography, and to become more pronounced as the climate deteriorated. A certain parallelism between the true Neandertal characters and the signs of rickets, thus, is not unexpected.

In spite of rickets and arthritis, the morphology of Neandertal man can be clearly recognized and interpreted as a harmonious functional adaptation to the environment in which he lived.

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Effect of Ethoxyquin on the Longevity of C3H Mice

SEVERAL WORKERS have found that mice fed diets containing large amounts of added antioxidant live longer than those fed standard laboratory diets. The size of the effect varies with the substance added, but it has been obtained with a wide variety of agents, including α -tocopherol, 2-mercaptoethylamine, ethoxyquin, *tert*-butyl hydroxytoluene (BHT), dithiocarbamates^{1,2} and (in rats) nor-dihydroguaiaretic acid³. These studies were prompted by the suggestion of Harman⁴ and of other workers^{5,6} that both natural and radiation-induced ageing may involve oxidative free radical attack on long term molecules or on lipids⁶, including those of mitochondria^{7,8}.

In the course of antioxidant screening studies we have observed a lifespan effect in C3H mice of both sexes receiving ethoxyquin in a standard pellet diet. Forty male and forty female 3 month old C3H mice, bred at the National Institute of Medical Research, were assigned randomly in equal numbers to control and antioxidant-supplemented groups, caged by tens with sexes separated, and kept in life table conditions, that is weighed monthly but subjected to no other manipulations.

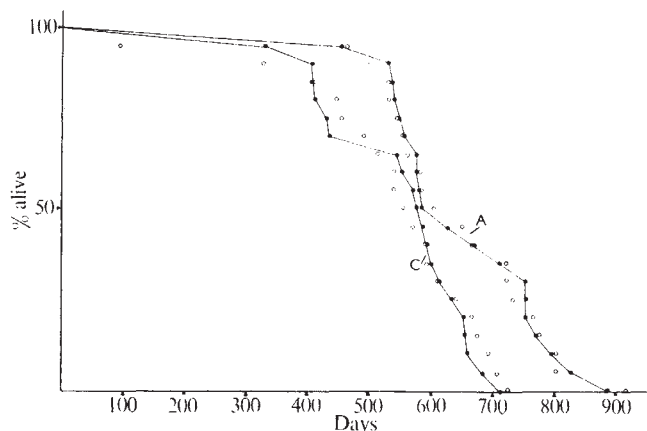


Fig. 1 Survival curves for mice on diet containing antioxidant (A) and on control diet (C). ●, Males; ○, females.

Control mice received a standard pellet diet ('Oxoid 41 B'), and treated mice received a diet of identical pellets moistened and reconstituted with 0.5% w/w ethoxyquin oil ('Santoquin', Monsanto), both given freely.

There was no sex difference in survival in either group, but survival curves for both sexes differed significantly in favour of the treated groups ($P < 0.005$, on a ranking test computed by Mr Peter Fayers, of the MRC Statistical Unit) with a curve displacement of about 100 days (Fig. 1). Tumour incidence in both treated and untreated groups seemed to be low for the strain. Mean expectations of life with standard errors and limit are given in Table 1.

Table 1 Longevity of C3H Mice on Standard Diet and Diet plus 0.5% w/w Ethoxyquin

	Control			Ethoxyquin	
♂	551.6 ± 24.7	(711)	♂	651.5 ± 27.3	(887)
♀	541.4 ± 33.3	(722)	♀	649.4 ± 27.4	(907)

Values are mean, s.e. and limit (days). n=Twenty per group.

Mice of both sexes fed antioxidant grew into significantly lighter adults than did controls, and lost weight earlier (Table 2). Activity and condition were strikingly better retained with age in the treated series. Pathological studies on a parallel series will be reported later.

Increased longevity in mice fed antioxidant is compatible with, but does not demonstrate, the hypothesis of free radical ageing. In the converse experiment, Gerschman⁹ found a decrease in mouse lifespan when oxygen tension was greater than 1 atmosphere, the effects being reversed by the administration of diethyl dithiocarbamate or Co^{2+} (ref. 10). In acute experiments, mice deficient in α -tocopherol increased in sensitivity to oxygen^{11,12}. Sobel¹³ failed to shorten life by intermittent exposure to 1.08 atmosphere of oxygen during youth and early adulthood.

In the antioxidant feeding experiments so far reported, alternative explanations are at least equally likely. None of the published survival curves, including our own, were obtained in a special purpose lifespan colony. The maximum lifespans obtained with antioxidants are within the recorded natural performance of the strains, and the increments are smaller than those regularly obtainable by calorie restriction.

Among alternative explanations, it could be suggested (1) that large amounts of chemicals hinder assimilation or spoil the appetite of mice, in the absence of strict pair-feeding measurements, and the effect may be due to covert calorie restriction; (2) that for mice, as for broiler fowls^{14,15}, excess antioxidant merely reduces the toxicity of a "normal" laboratory diet²; (3) that (according to unpublished studies by R. B. Cumming) many antioxidants, especially BHT, are powerful enzyme

Table 2 Weights of C3H Mice on Control (C) and Antioxidant-containing (A) Diets (Gm)

	97	125	197	248	310	409	533
♂							
C	25.4±1.86 (22-30) n=20	27.7±2.62 (23-31) 20	31.9±2.84 (29-36) 20	32.6±2.64 (29-34) 20	30.5±1.73 (26-33) 20	30.5±2.12 (26-32) 19	29.2±3.04 (26-31) 13
A	26.0±1.79 (23-29) n=20	28.3±2.11 (25-34) 20	28.9±2.29 (25-36) 20	27.9±2.44 (24-34) 20	25.2±2.72 (22-29) 20	26.3±2.87 (22-30) 20	26.0±3.04 (22-36) 19
♀							
C	20.2±1.30 (18-23) n=20	22.1±1.70 (20-25) 20	26.8±2.59 (22-31) 19	28.7±3.15 (24-33) 19	27.3±3.03 (23-33) 19	26.6±3.45 (21-31) 18	25.9±2.99 (25-32) 13
A	20.9±2.27 (16-28) n=20	22.3±1.78 (20-28) 20	24.4±2.37 (21-29) 20	24.1±2.05 (22-28) 20	20.6±2.04 (18-25) 20	21.0±2.51 (17-25) 20	20.3±1.78 (17-24) 18

Values are mean, s.d. and range.

inducers—ethoxyquin in these doses causing marked liver enlargement¹⁶: in Ross's experiments, hepatic enzyme concentrations were found to correlate strongly with further rat life expectancy on various diets¹⁷; (4) that with the doses used a hypothesis of straightforward "chemical stress", with or without suppression of a predominant tumour, is a feasible cause of longer gross survival. None of these possibilities has been excluded. Experiments with other potent enzyme inducers, such as DDT or barbiturates, seem to be necessary.

The form of the curves, and their response to a mortality incident affecting all groups after between 500 and 600 days, which was probably infective or environmental, would be consistent with a postponement of some predominant age-dependent process by about 15%, rather than with the suppression of one incidental cause of senile mortality. Non-actuarial indices of ageing (pigment deposition, tumour incidence) remain to be examined and will be reported later. We wish, however, to confirm the existence of the effect in a strain previously reported to be unaffected by antioxidants¹⁸.

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Stimulation by Artificial Lighting of Calcium Absorption in Elderly Human Subjects

VITAMIN D, the "sunshine vitamin", is produced in the skin when ultraviolet radiation is absorbed by the pro-vitamin 7-dehydrocholesterol¹. The endogenous vitamin, together with dietary vitamin D, is then apparently converted in the liver to 25-OH vitamin D, and this active metabolite facilitates the intestinal absorption of calcium and the uptake and release of calcium by the skeleton^{2,3}.

The amounts of vitamin D usually produced in the intact human skin are unknown. Ultraviolet irradiation of the skin cures childhood rickets⁴⁻⁷; in this case endogenous production must be at least equivalent to the minimum curative dietary dose of 200-400 iu daily⁸. Extrapolation of these results to adults is difficult, because their dietary requirement for vitamin D is not established⁸. But there is indirect evidence, summarized by Loomis⁹, that the production of vitamin D induced by ultraviolet light is important in the skin.

Ordinary window glass absorbs essentially all radiation of the wavelength necessary for this *in vivo* synthesis—between 275 and 310 nm—of vitamin D⁴⁻⁷. Millions of people work behind glass, underground or in the extreme north, travel to and from work in closed vehicles, and venture outdoors only in the early morning or late evening, when ultraviolet radiation is minimal¹⁰. Incandescent bulbs emit little ultraviolet radiation; the small amount from ordinary fluorescent bulbs is usually absorbed by the fixtures in which they are mounted. We have examined changes in calcium absorption after exposure to 'Vita-Lite' (Duro-Test Corporation, North Bergen, New Jersey), a commercial fluorescent lamp of conventional geometry and loading, designed to duplicate Sun and sky radiation at a colour temperature of 5,500 K (CIE D-5500). About 5% of its total radiant power lies at wavelengths between 290 and 380 nm. Our data suggest that illumination which simulates natural light significantly increases the efficiency of intestinal calcium absorption in people who receive no ultraviolet light from the Sun.

Eighteen white, male residents of the Chelsea Massachusetts Soldiers' Home were selected on the basis of ability to cooperate, age (57-80) and freedom from significant disease. All studies were done with their voluntary consent.

From December 20, 1968, to April 25, 1969, subjects were asked to stay indoors and away from open windows during daylight. Men who normally took multivitamin preparations containing vitamin D were given a substitute lacking this vitamin. Their normal diet contained restricted types of seafoods. They were asked to maintain their habitual intakes

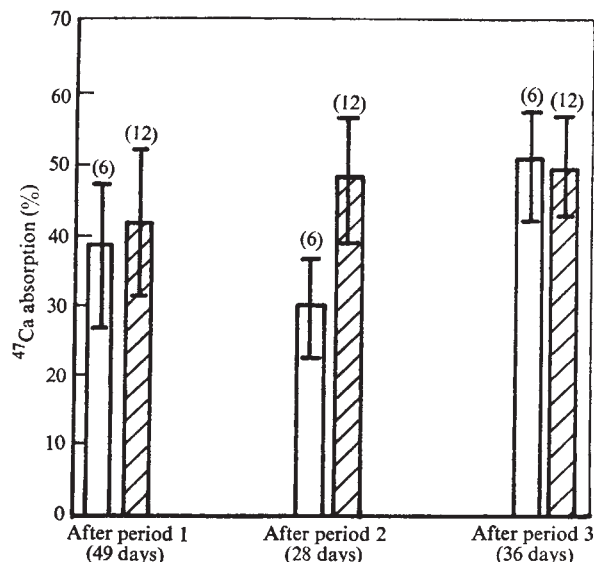


Fig. 1 Absorption of ^{47}Ca among six control and twelve experimental subjects during the three periods of the study (means $\pm$ s.e.). The experimental group was exposed to 'Vita-Lite' during period 2. White columns, control group; hatched columns, experimental group.

of dairy products, which provided an estimated 200 IU or less of vitamin D daily. Their estimated calcium intake throughout the study was 500–1,000 mg daily, the recommended normal intake¹¹. Subjects who habitually consumed less than this were given daily calcium supplements.

The study was divided into three periods: period 1 (December 20, 1968–February 7, 1969) was used to allow endogenous vitamin D and 25-OH vitamin D stores to stabilize at new levels consistent with the changes in routine. The time needed to attain this steady state was unknown; 7 weeks were allowed arbitrarily. During this time, subjects were exposed to the low level of ambient lighting (10–50 foot candles of incandescent and/or fluorescent) usually present within the Chelsea Home. At the end of period 1, the ability of each subject to absorb calcium was estimated by measuring the percentage of ^{47}Ca passed in the faeces within 6 days of ingestion. A complete metabolic research ward was established at the home for all data collection periods. One microcurie of $^{47}\text{CaCl}_2$ in 4 fluid ounces of milk was given 2 h before breakfast after an overnight fast. Complete 6 day faecal collections were homogenized, and 1,500 ml. aliquots were counted with appropriate standards to a precision of $\pm 1.5\%$ in an 'Atomium GD-1' detector, using a lower discriminator of 0.5 meV. Percentage absorption was 100% minus the percentage excreted, without correction for recycling, when such small doses of ^{47}Ca are used. In all studies less than 1% of the ^{47}Ca dose was excreted in the specimen collected on day 6. During period 2 (February 15, 1969–March 15, 1969), experimental subjects were exposed to the test lights ('Vita-Lite' in ultraviolet-reflective fixtures, 500 foot candles) for 8 h/day. Control subjects were similarly exposed to cool-white fluorescent bulbs in standard luminaries (30–50 foot candles) representing their normal light environment. For the remainder of their waking hours, all subjects lived under the same ambient lighting as during period 1. At the end of this period, ^{47}Ca absorption was again measured. In period 3 (March 21, 1969–April 25, 1969) all subjects were exposed to the same conditions of artificial light as during period 1. Statistical analyses were made using the paired *t* test for intragroup changes and the unpaired *t* test for intergroup changes.

At the end of period 1 ^{47}Ca absorption was similar in control and experimental groups ($39 \pm 9\%$ and $41 \pm 10\%$ mean

$\pm$ s.d.). The ranges (23–54%) were those reported for other normal subjects^{12–14}.

During period 2, mean, ^{47}Ca absorption in the control group decreased to $30 \pm 8\%$, but increased to $47 \pm 9\%$ in the men exposed to the test lights ($P < 0.01$). Serum and urinary calcium were unchanged. The decline in the control group probably reflects a continuing decrease in body vitamin D activity resulting from dietary changes and the unavailability of ultraviolet radiation. This suggests that period 1 (49 days) was not long enough to attain a new steady state in calcium absorption. The increased ^{47}Ca absorption in men exposed to test lights suggests that the small amount of ultraviolet radiation from these lights was sufficient to stimulate significant vitamin D synthesis in the skin, not only preventing the continued depletion of the vitamin, but enhancing the body supply. The changes in vitamin D or 25-OH vitamin D needed to account for such results are unclear, for the dose-response curve relating vitamin D or 25-OH vitamin D to intestinal ^{47}Ca absorption is undefined in man.

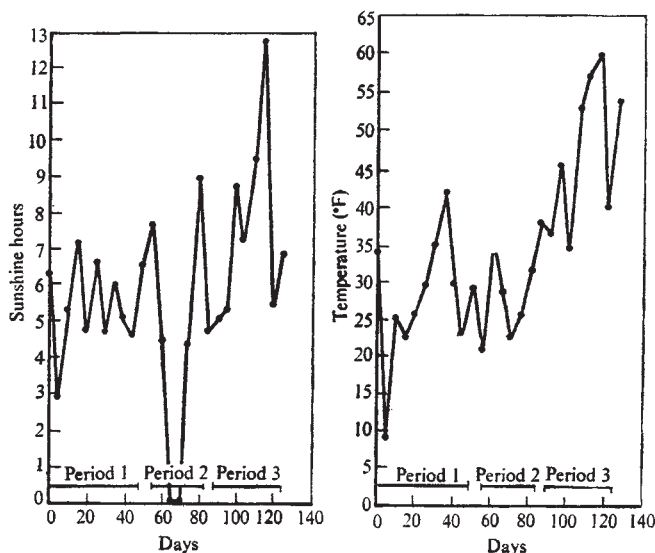


Fig. 2 Sunshine hours and average temperature plotted every fifth day throughout the period December 20, 1968, to April 25, 1969.

The effect of the test lights may be unrelated to their ultraviolet emission or to increased vitamin D synthesis. 'Vita-Lite' resembles natural Sun and sky radiation and differs from standard cool-white fluorescent lighting in not only the ultraviolet but also the visible emission. The intensity of the visible radiation was also much greater for the test group (500 foot candles) than for the control subjects (30–50 foot candles) during period 2. The neuroendocrine apparatus is sensitive to such differences in lighting^{15–20}, and so it is possible, if unlikely, that variations in the visible portion of the spectrum mediated the differences in calcium absorption.

By the end of period 3, ^{47}Ca absorption in the control group had also increased significantly ($51 \pm 7\%$; $P < 0.01$ intra-group) while in the experimental subjects it was virtually unchanged ($49 \pm 7\%$). Absorption of ^{47}Ca in both the control ($P < 0.01$) and experimental ($P < 0.05$) groups was significantly higher at the end of period 3 than at the end of period 1. The changes in the control group during period 3 cannot be attributed to the artificial lighting, which was the same as in period 1. Further studies will be necessary to define the uncontrolled variable during period 3, but one of the leading possibilities is external illumination. During period 3, the weather in Boston was considerably sunnier than during the first two periods (Fig. 2). Moreover, the average amount of

ultraviolet energy per hour of noonday sunlight reaching Boston during period 3 was three to five times as great as during periods 1 and 2 (refs. 10, 21). At least one man went outside; others sat looking southward through windows which were sometimes open. It seems likely that these men received significant doses of natural ultraviolet light.

These results suggest that relatively small amounts of ultraviolet light can stimulate calcium absorption among elderly men who have no exposure to sunlight and eat a diet containing few foods fortified with vitamin D. Further studies are necessary to confirm these findings and to define the population groups to which they may apply. It is possible that the ability to absorb dietary calcium might be enhanced in large sections of the population by exposure to small amounts of ultraviolet light provided by the general lighting system. The amounts involved seem to be roughly equivalent to those that would impinge on a resident of Washington DC who took a daily 15 min walk out of doors at lunchtime in midsummer.

The changes which occurred between period 1 and period 3 could be construed as evidence that there is seasonal variation in calcium absorption and perhaps vitamin D synthesis, at least among individuals who are exposed to little sunlight.

We thank Mr Sergei Marketan, Duro-Test Corp., North Bergen, New Jersey, who designed and installed the lighting systems, and Dr James Coyle and the staff of the Chelsea Soldiers' Home for their help and cooperation.

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Visna Virus: a Slow Virus with an RNA Dependent DNA Polymerase

VISNA virus is the cause of a progressive neurological disease of sheep which results in paralysis and death^{1,2}. After inoculation, the virus multiplies within the host, but illness may not be manifest for months or years³. Visna virus shares some physical, chemical and biological properties with oncogenic RNA viruses⁴. The similarities include virion and core size, site of maturation, growth properties, nucleic acid composition⁵, and sensitivity to physical and chemical agents. The inhibition of visna virus replication by low concentrations of actinomycin D (ref. 5) indicates a requirement for DNA synthesis for the growth of this agent. An RNA dependent DNA polymerase has been discovered in many oncogenic RNA viruses^{6,7}. This communication demonstrates the presence of similar enzyme activity in visna, a virus without known oncogenic potential.

Visna virus and sheep anti-visna serum were provided by Dr Halldor Thormar of the Institute for Basic Research, Staten Island. The virus was plaque purified and propagated in a sheep testes cell strain. Neutralization by the sheep anti-serum established its identity. Supernatant culture fluids from visna-infected cell monolayers were clarified by low speed centrifugation and concentrated with 10% polyethylene glycol (Union Carbide, PEG-6000) in the presence of 0.5 M NaCl. This material was then either pelleted through 10% (w/v) sucrose or purified by banding in a preformed continuous gradient of 20–70% (w/v) sucrose for 18 h at 257,000g in an SW-65 rotor. The final titre of visna virus used in polymerase assays averaged 10⁸ TCID₅₀/ml. The virus used in these experiments was free from contaminating mycoplasma and known murine oncogenic RNA viruses.

Table 1 Requirements for RNA Dependent DNA Polymerase in Visna Virus

	Apmol ³ H-TTP incorporated
Complete	0.50
+ RNase A (1.0 mg/ml.)	0.20
– dATP	0.02
– dATP, dGTP, dCTP	0.02
+ Poly rA.rU	4.10
+ Poly rA.rU – dATP, dCTP, dGTP	4.05
– Virus	0.02
– Virus + poly rA.rU	0.02
– DTT	0.02
– Mg ²⁺	0.02
– 'Triton'	<0.01

Each reaction mixture was incubated for 90 min at 37° C and contained in the complete system in 0.05 ml.: 0.04 M Tris HCl (pH 7.8); 0.06 M potassium chloride; 0.01 M magnesium acetate; 5 × 10^{–4} M dATP, dCTP, dGTP; 2 × 10^{–5} M ³H-methylthymidine triphosphate (6,000 c.p.m./pmol); 8 × 10^{–3} M dithiothreitol; 0.22 A₂₆₀ poly rA.rU where indicated; 0.1% 'Triton X-100' (% v/v) and 10⁶ TCID₅₀ of virus. RNase A was heated for 15 min at 90° C to inactivate any possible contaminating DNase. The zero time value in the complete system of 0.07 pmol has been subtracted from all values. ³H-methylthymidine triphosphate was purchased from Schwarz BioResearch; poly rA.rU and deoxynucleoside triphosphates from Miles; RNase A from Calbiochem; DNA synthesis was assayed as previously described^{6,7}.

Table 1 shows the requirements for the visna virus DNA polymerase. A seven-fold increase in the incorporation of ³H-TTP into DNA over the zero time value can be seen. Maximal DNA synthesis required all four deoxynucleoside triphosphates, a divalent cation, 'Triton X-100', and a sulphhydryl agent. DNA synthesis was inhibited by RNase A treatment of detergent-disrupted virus suggesting that the template was RNA.

These findings are similar to those observed with the RNA dependent DNA polymerase of C-type oncogenic viruses^{6,7}. Recently synthetic RNA polymers have been used to stimulate DNA polymerase activity in C-type oncogenic viruses⁸. Table 1 also shows that one of these templates, poly rA.rU, was

able to stimulate the incorporation of ^3H -methyl-TTP into DNA with the visna virus polymerase. Incorporation did not require dGTP, dCTP, or dATP, a result also found with C-type RNA virus polymerases⁸.

Earlier studies showed a critical dependence on detergent concentration for demonstration of DNA polymerase activity in RNA tumour viruses^{6,7}. Table 2 shows the effect of different 'Triton X-100' concentrations on visna virus polymerase activity. Maximal synthesis was observed at a concentration of 0.1% 'Triton X-100', a somewhat higher optimum than that observed for RNA-containing tumour viruses⁹. Incorporation was not observed at any concentration of detergent with the omission of one deoxyribonucleoside triphosphate. No incorporation into RNA could be detected when the four riboside triphosphates were substituted for the DNA precursors.

Table 2 'Triton' Dependence for DNA Polymerase Activity in Visna Virus

'Triton' (% v/v)	Δ pmol ^3H -TTP incorporated -dGTP	+dGTP
0.000	<0.01	0.02
0.006	—	0.02
0.014	<0.01	0.17
0.028	<0.01	0.60
0.040	—	0.62
0.100	—	0.65
0.200	<0.01	0.60
0.400	—	0.42

Each reaction mixture was incubated for 90 min at 37° C and contained components as listed in Table 1 except for 'Triton' as indicated. The zero time value of 0.07 pmol has been subtracted from all values.

The demonstration of a visna virus DNA polymerase provides additional evidence of the similarities between visna and the avian and murine leukaemia viruses previously noted by Thormar⁴. The biological function of the visna polymerase is unknown. Oncogenic RNA viruses have been reported to integrate as a DNA copy into the host cell genome^{10,11}. The RNA dependent DNA polymerase of oncogenic RNA viruses is presumably involved in the integration mechanism. The DNA polymerase of the RNA virus, visna, might serve a similar function. How integration of visna virus into the host cell genome might be related to the pathogenesis of the visna disease state is under investigation.

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RNA Dependent DNA Polymerase in Primate Syncytium-forming (Foamy) Viruses

SINCE the initial observations of Enders and Peebles¹ and Rustigian *et al.*², syncytium-forming ('foamy') viruses have frequently been isolated from monkey kidney cultures and are regarded as 'common contaminants' of these cells. Infection by these agents often leads to the formation of multinucleated giant cells (syncytia). Viral replication involves intracytoplasmic formation of a complete nucleoid, and the outer membrane is acquired from either the plasma membrane or the endoplasmic reticulum^{3,4}. In their morphology and mode of replication, syncytium-forming viruses are similar in some ways to the RNA-containing tumour viruses⁵. We wish to show that the primate syncytium-forming viruses contain RNA and, like the RNA-containing tumour viruses, replicate most efficiently in dividing cells. Further, viral replication can be inhibited by small doses of actinomycin D. We shall demonstrate the presence of an RNA dependent DNA polymerase in purified virions.

In the course of other studies, a syncytium-forming virus was isolated from uninfected African green monkey kidney cultures (GMK). Neutralization tests indicated that the virus was a strain of foamy virus type 3⁶. In certain cell cultures, syncytium formation was detectable within 48 h of infection. The rapidity with which this process occurred suggested fusion of cell membranes of adjacent cells as the mechanism for syncytium formation. Syncytia were observed much later and with greatly reduced frequency in cultures that were confluent when infected. 'Latent' viruses, however, could be detected when such cultures were serially transferred. The cytopathic effects progressed, sometimes to the destruction of the entire culture. Cells of several species supported the growth of the virus (Table 1). Rat cells grew the foamy virus to the highest titre, and were the most sensitive of the cultures assayed for syncytium formation.

Table 1 Host Range of Primate Syncytium-forming (Foamy) Virus Type 3

Species	Cell	Infectivity range *
Human	HK	10 ³ -10 ⁴
	WI-38	n.d.
Simian	GMK	10 ³ -10 ⁴
	CV-1	10 ⁴
Mouse	NIH/3T3	10 ⁴
	BALB/3T3	10 ³ -10 ⁴
Rat	NRK	10 ⁴ -10 ⁵

* Syncytium-forming units/ml.

Clarified virus from supernatants of infected GMK cells was incubated with monolayer cultures for 1-2 h at 37° C with frequent rotation. Medium was replaced and infectivity was determined by counting syncytia at each virus dilution. HK, Human kidney; WI-38, human embryonic lung strain; GMK, green monkey kidney; CV-1, GMK heteroploid cell line; NIH/3T3 and BALB/3T3, mouse cell lines⁷; NRK, normal rat kidney⁸. n.d., Not done.

When cultures in which about 50% of the cells were involved in syncytia were incubated with ^3H -uridine (New England Nuclear Corporation; specific activity > 20 Ci/mmol) for 16 h and the concentrated supernatants were sedimented in a linear 15-55% sucrose gradient, a distinct peak of radioactivity was seen at 1.16-1.17 g/cm³. A smaller peak of radioactivity at a density of 1.22-1.24 g/cm³ may represent the viral nucleoid⁹. Parallel cultures incubated with ^3H -thymidine showed no detectable peak. Biological activity was maximal at a sucrose density of 1.16 g/cm³ when gradient fractions were tested for infectivity on GMK cells.

When virus-infected cultures were incubated with actinomycin D in a concentration of 1 µg/ml. for 6 h before labelling with ^3H -uridine, the peak of radioactivity was abolished (Fig. 2). The yield of syncytium-forming virus was also decreased from

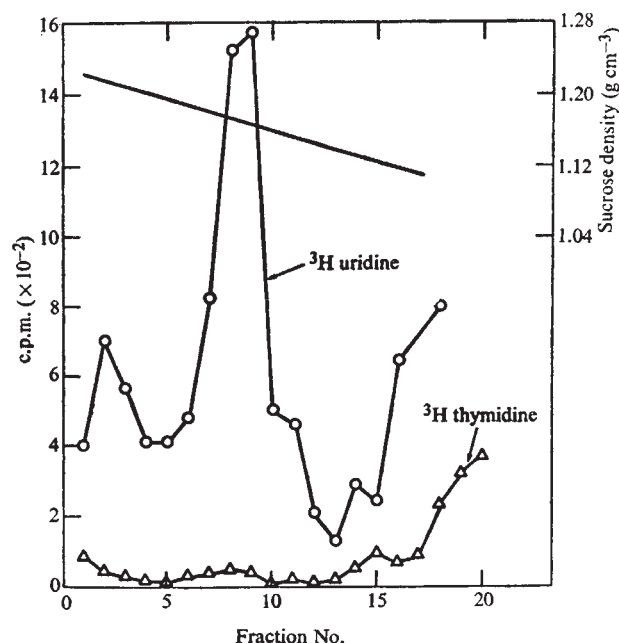


Fig. 1 Incorporation of ^3H -uridine or ^3H -thymidine ($30 \mu\text{Ci}/\text{ml.}$) into GMK cultures infected with primate syncytium-forming virus. Cultures were incubated with the labelled compounds for 16 h. Supernatant fluids were clarified by centrifugation at $2,000g$ for 10 min, then precipitated with saturated $(\text{NH}_4)_2\text{SO}_4$. The precipitate was redissolved in 1.5 ml. of buffer, layered over a 15–55% linear sucrose gradient and centrifuged at $200,000g$ for 2 h. Fractions were collected, precipitated with 10% trichloroacetic acid (TCA), collected on filters, and washed twice with 5% TCA. Filters were counted in Bray's scintillation fluid in a liquid scintillation counter.

a titre of 5×10^3 to 4.5×10^2 syncytium-forming units per ml. after a 24 h exposure of infected cells to $1 \mu\text{g}/\text{ml.}$ of actinomycin D. Parallel cultures of GMK cells infected with vesicular stomatitis virus (VSV), which contains an RNA dependent RNA polymerase and does not require DNA synthesis for its replication¹⁰, showed no decrease in the incorporation of radioactivity with this concentration of actinomycin D.

Supernatant fluids from infected cultures showing advanced cytopathic effect were clarified and concentrated thirty-fold by ultracentrifugation. The concentrates were then assayed for RNA dependent DNA polymerase^{11,12}. The requirements for primate syncytium-forming virus-stimulated DNA synthesis

Table 2 Requirements for DNA Polymerase Activity

Complete	$\Delta\text{pmol } ^3\text{H-TMP}$ incorporated
—dATP	0.30
—dATP, dGTP, dCTP	0.01
— Mn^{2+}	< 0.01
— $\text{Mn}^{2+} + \text{Mg}^{2+}$	0.26
—virus	< 0.01
—'Triton X-100'	< 0.01

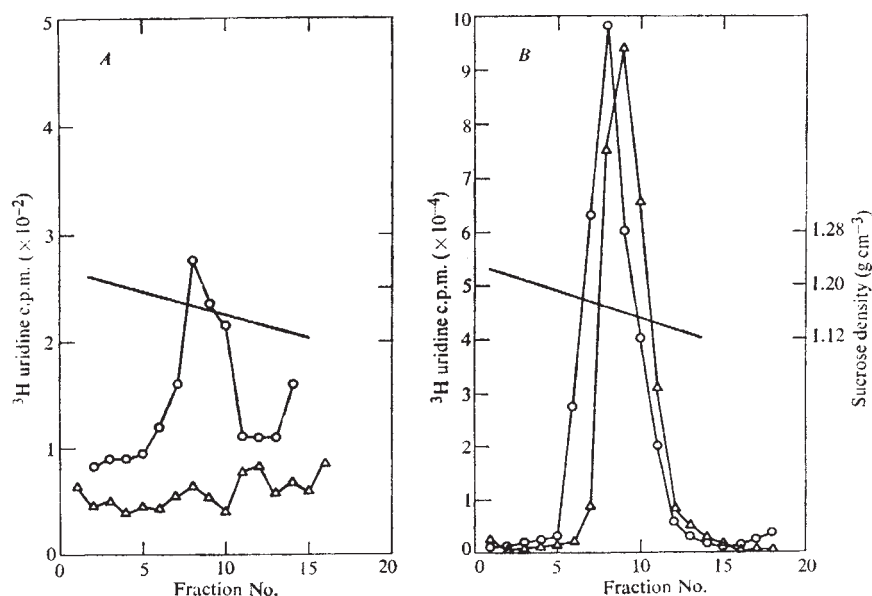
Each complete reaction mixture was incubated for 90 min at 37°C and contained in 0.05 ml: 0.04 M Tris HCl (pH 7.8); 0.06 M potassium chloride; 0.002 M manganese acetate; 0.002 M dithiothreitol (DTT); 5×10^{-4} M dATP, dGTP, dCTP, and 2×10^{-5} M ^3H -methyl-TTP; 5.0 μg of foamy virus protein; 0.2% (v/v) 'Triton X-100'. The zero time value of 0.10 pmol was subtracted from each determination.

are shown in Table 2. DNA synthesis required all four deoxyribonucleoside triphosphates, a divalent cation, and 'Triton X-100'. No incorporation was found with the four ribonucleoside triphosphates. In experiments not shown, maximum incorporation was obtained with 0.1–0.2% 'Triton X-100'. This relatively high requirement for detergent is different from the optimum concentration (0.014%) for detecting polymerase activity in the murine leukaemia viruses¹³.

Earlier studies had shown that synthetic nucleic acid templates could markedly increase the sensitivity of detection of polymerase¹⁴. Table 3 shows the effects of adding three synthetic polymers to the primate virus enzyme system. Incorporation of radioactivity by disrupted virions was markedly increased with the synthetic RNA copolymer, rA.rU, the synthetic DNA complex, dAT, and a synthetic RNA–DNA copolymer, dC.rG. Thus, foamy virus contains enzyme(s) capable of transcribing double stranded RNA, double stranded DNA, and an RNA–DNA hybrid. The pattern of relative activities observed with the different copolymers is similar to that found in the murine leukaemia virus enzyme system. Polymerase activity was also found in virions after passage through human or rat cells. Prototype foamy virus type 2 (titre $7.3 \text{ TCID}_{50}/\text{ml.}$) and type 3, obtained from Dr R. Holdenreid (Resources and Logistics Segment, NCI), also had DNA polymerase activity. No enzyme activity has been detected in concentrated supernatants from uninfected GMK cells or VSV-infected GMK cells.

Earlier work on murine leukaemia virus RNA dependent DNA polymerase activity with partially disrupted virus had suggested that enzyme activity could be recovered from sucrose

Fig. 2 Effect of actinomycin D on ^3H -uridine incorporation into primate syncytium-forming virus (A) and vesicular stomatitis virus (B). GMK cultures were treated with $1 \mu\text{g}/\text{ml.}$ of actinomycin D 6 h before addition of the radioactive label. Twelve hours later the supernatant fluids were harvested and processed as in the legend to Fig. 1.



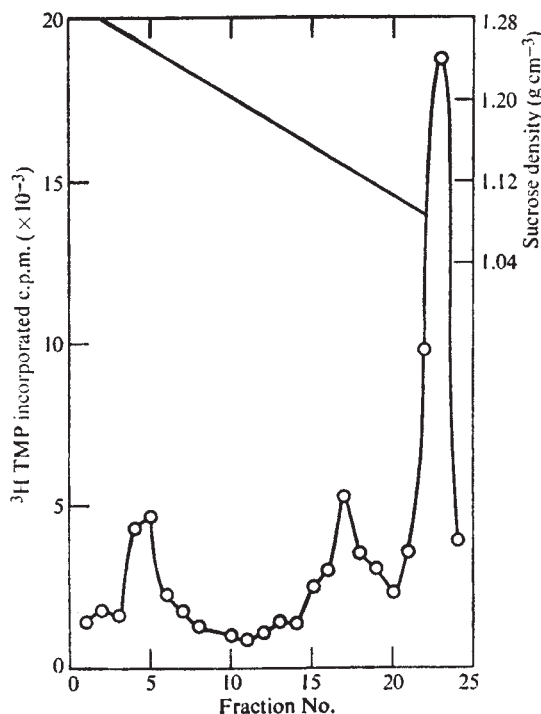


Fig. 3 Supernatant fluids from infected GMK cells were concentrated thirty-fold by pelleting at 75,000g for 2 h, resuspended in buffer and layered over a linear 15–60% sucrose gradient. After centrifugation at 200,000g in an SW-41 rotor for 2 h, fractions were collected by bottom puncture and assayed directly for polymerase activity. Each reaction was incubated at 37° C for 90 min and contained in 0.05 ml.: 0.04 Tris HCl (pH 7.8); 0.05 M potassium chloride; 0.001 M manganese acetate; 0.002 M DTT; 2×10^{-5} M ^3H -methyl-TTP; 0.2% Triton X-100; 0.006 A_{260} rA.rU; and 0.02 ml. of each fraction.

gradients in a viral fraction banding at 1.14–1.16 g/cm³, in a viral core at 1.24 g/cm³ and in a “soluble” fraction at 1.08 g/cm³ (ref. 15). When concentrated foamy virus preparations were layered on sucrose gradients and the fractions were assayed for enzyme activity, three clearly separated peaks of activity (at sucrose densities of 1.22, 1.14 and 1.08 g/cm³) were demonstrated, using the synthetic polymer, rA.rU (Fig. 3). The endogenous reaction (in the absence of the added polymer) was considerably lower and could only be detected in the virion and nucleoid fractions.

Electron microscopy of various gradient fractions revealed viral core material in the 1.22–1.24 g/cm³ region, intact viral particles in the 1.16 g/cm³ region, and only damaged virions in

the 1.14 g/cm³ region of the gradient. The lightest fractions contained only what seemed to be virion debris. Viral infectivity was recoverable at low titre but only from the 1.16 g/cm³ region.

RNA dependent DNA polymerases are not known to be associated with tumours or with any other diseases; they can be recovered easily from normal monkey kidney cells¹⁶. Another “non-oncogenic” virus, visna, which is the cause of a progressive neurological disease of sheep^{17,18}, has been shown to contain similar enzyme activity. These observations suggest two alternatives. The first is that these viruses as well as visna virus may be oncogenic. With neither of these agents has this possibility been tested adequately, although Clarke *et al.* noted morphological alterations in BHK-21 cells persistently infected with foamy virus type 1¹⁹. The second possibility is that the presence of an enzyme that transcribes RNA into DNA may be a prerequisite for RNA-containing viruses to become latent in cells.

Several other recently described syncytium-forming viruses of avian, feline and bovine origin have many characteristics that closely resemble those of the primate syncytium-forming viruses^{20–23}; they have been observed in tumours of several species^{20,22,23}. For example, a virus recently isolated from a spontaneous rhesus monkey mammary tumour has the morphological characteristics of the primate syncytium-forming “foamy” viruses²⁴. A re-examination of the role of this long-neglected group of viruses in oncogenesis and chronic diseases is warranted.

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Table 3 Comparison of Polymer-stimulated DNA Synthesis

Polymer additions	Δ pmol incorporated
	^3H -TMP
None*	0.30
rA.rU	3.69
dAT	19.80
	^3H -GMP
dC.rG	1.69

* Conditions were as described in Table 2.

Each reaction mixture was incubated for 90 min at 37° C and contained in 0.05 ml.: 0.04 M Tris HCl (pH 7.8); 0.06 M potassium chloride; 0.002 M manganese acetate; 0.002 M DTT; 0.1% (v/v) Triton X-100; 2×10^{-5} M ^3H -methyl-TTP (6,600 c.p.m./pmol) and 1×10^{-4} M dATP or 2×10^{-5} M ^3H -dGTP; 5 μg of foamy virus protein. Incorporation minus polymer of 0.10 pmol of ^3H -TMP or 0.08 pmol ^3H -dGMP was subtracted from the appropriate polymer reaction. The amount of polymer used was: rA.rU 0.006 A_{260} ; dAT 0.03 A_{260} ; dC.rG 0.0003 A_{260} . Poly rA.rU is the double stranded complex of the two homopolymers of polyriboadenylic acid and polyribouridylic acid; dAT is the double stranded copolymer composed of alternating dA and dT units. dC.rG is the double stranded complex of the two homopolymers polydeoxycytidylic acid and polyriboguanidylic acid.

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Concentrations of Plasma Testosterone in Normal Men during Sleep

IN man, several studies^{1,2} have demonstrated diurnal variation, with a morning peak, in plasma testosterone—the most potent naturally occurring androgen. The origin of this variation is not known.

Diurnal changes and fluctuations in circulating concentrations of other hormones have, however, been studied more extensively. Concentrations of plasma corticosteroid also have a morning peak; during sleep they remain low, until between 0400 h and 0800 h they reach a series of successive peaks which create the morning rise³. Recent studies have confirmed that it is possible to dissociate these peaks from sleep⁴. Concentrations of growth hormone in plasma have, on the contrary, been shown to be closely associated with a component of normal sleep⁵, and an increase in the circulating concentration of this hormone persists when the subject sleeps during the day. The serious disturbance of the diurnal variation of testosterone when routines of sleep and wakefulness were disorganized⁷ suggested that the overnight concentrations of testosterone were linked to sleep.

Two separate and distinct physiological states alternate regularly throughout the sleep period. A cycle of orthodox or slow wave sleep which is characterized by slow waves and sleep spindles in the electroencephalogram (EEG), regularity of most physiological variables and relative absence of mental activity, occupies the first 60–90 min. It is succeeded by a period of “paradoxical” or rapid eye movement (REM) sleep in which large jerking eye movements occur against a lower voltage EEG background. Irregularity of physiological variables is striking and reports of dream activity frequently occur retrospectively. A cycle of slow wave sleep and a period of REM sleep occupy a mean of 90 min and REM sleep constitutes a mean of 24% of total sleep in adults.

The relationship of testosterone to sleep has been examined in two experiments. Five healthy male adult subjects (aged between 19 and 45) attended the sleep laboratory at intervals during a 4 month period. Electrodes were attached to scalp,

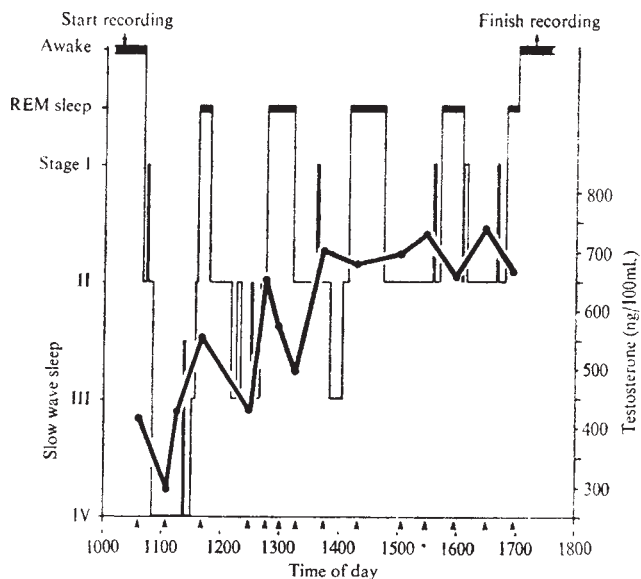


Fig. 2 Diurnal sleep. There were five cycles of slow wave sleep with five interdigitating periods of REM sleep. Plasma testosterone decreased during the initial slow wave sleep cycle, and thereafter there were successive peaks before or at the start of each period of REM sleep which produced an overall increase. This effectively reversed the normal diurnal decrease in plasma testosterone.

around the eyes and on the neck as described elsewhere⁸, so that eye movements, EEG, muscle tone and pulse rate could be monitored continuously overnight. A Deseret catheter was inserted into a forearm vein and filled with dilute heparin/saline solution. This was connected to a 10 foot nylon catheter extension so that samples could be removed remotely during sleep⁹. Samples taken at a mean interval of 30 min were placed in lithium heparin tubes and centrifuged promptly. Plasma was pipetted off and deep frozen immediately. An average of eighteen samples were collected per subject night. The continuous recording of the physiological changes of sleep was analysed by internationally agreed criteria¹⁰.

Plasma testosterone concentrations were measured by the competitive protein binding method¹¹. The samples from each subject night were analysed as a group, in duplicate whenever possible. The technique is reproducible and is reasonably specific for testosterone; the coefficient of variation is about 12%. The lower limit of sensitivity is 1 ng/sample. Data from thirteen subject nights form the basis of this communication.

Although sampling was relatively infrequent, all subjects showed evidence of an overall increase in plasma testosterone on almost every night. The slope of the increase varied between subjects and within successive subject nights, although the trend remained. Fluctuations in plasma testosterone concentrations were superimposed on the general upward trend. A decline was usual during the initial cycle of slow wave sleep, after which successive peaks in the plasma concentration of hormone created a stepwise increase during the night. Occasionally samples taken close together revealed a dramatic increase (Fig. 1) which could only be explained by active secretion.

Peaks of testosterone occurred in conjunction with or adjacent to periods of REM sleep. Concentrations frequently increased before the period of REM sleep, and sometimes if the subject awoke when such a period was due, the increase in plasma testosterone was evident (Fig. 1). Arousal and sudden shifts towards arousal were also often associated with an increase in plasma testosterone. In one subject, sleep during the day was accompanied on several occasions by a typical increase in testosterone which reversed the normal diurnal trend (Fig. 2).

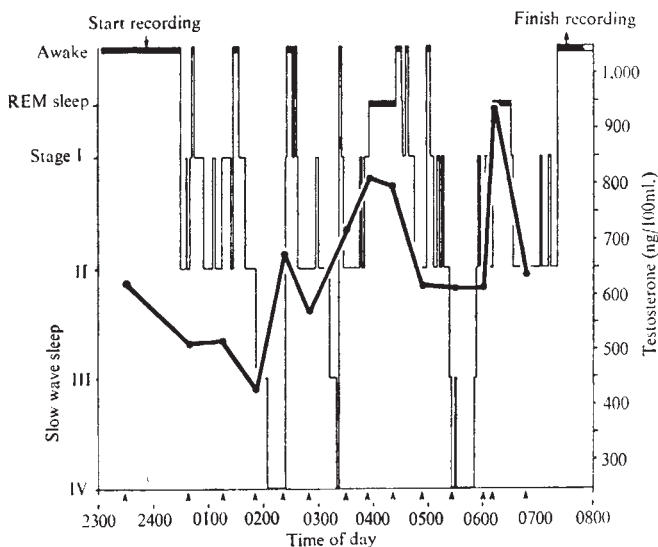


Fig. 1 Nocturnal sleep. During this night sleep, there are three principal cycles of slow wave sleep and two REM sleep periods. A peak of testosterone occurred at the onset of the third REM sleep period at 0600 h. Concentrations of testosterone were increasing before the second REM sleep period at approximately 0400 h. At approximately 0230 h, however, when a period of REM sleep was due, the subject awoke. The expected increase in plasma testosterone is still apparent but there was a decrease when the subject returned to sleep.

REM sleep clearly does not trigger the production of hormone, but the relationship suggests a link between the neurophysiological state underlying REM sleep and the mechanism regulating the production of testosterone. REM sleep is itself associated with penile tumescence and erection, and on occasion a frankly erotic dream may give rise to an emission during REM sleep¹². Alternatively, testosterone is an anabolic hormone and restorative or reparative roles have been suggested for REM sleep¹³.

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Adverse Influence of Ozone on Pulmonary Bactericidal Activity of Murine Lung

As an air pollutant, ozone is considered to be an important predisposing factor in pulmonary bacterial infections¹⁻². Little is known about the precise mechanisms responsible for the diminished resistance to infection, but it seems that the pathogenesis of these infections involves inhibition of pulmonary antibacterial activity. *In vivo* investigations have demonstrated that mice exposed to ozone have an increased mortality from infection with pathogenic streptococci³ or *K. pneumoniae*^{2,4}. Experiments using an *in vitro* method for measuring alveolar macrophage function have shown marked impairment in phagocytic activity after exposure of mice to ozone⁵. These results prompted us to study the effect of ozone on the *in vivo* rate of bacterial killing by the murine lung. We show inhibition of pulmonary bactericidal activity from short term exposures to ozone concentrations as low as 0.62 p.p.m.

The methods used for the *in vivo* determination of the antibacterial activity rate of the pulmonary phagocytic system have been published⁶. Forty or fifty male Swiss 25 g mice were infected with aerosols of ³²P-labelled *Staphylococcus aureus*. Ten animals (0 h) were killed with ether after infection and the radioisotope concentrations of their lungs measured. The rest of the infected animals were divided into groups of ten and exposed in fibreglass chambers for 4 h to the following

mean concentrations of ozone: 0.62, 0.70, 0.80, 1.10, 2.23, 2.28, 2.65 and 4.25. Transfer of animals from the infection chamber to the ozone chambers took 30–45 min. Ozone was generated from oxygen by a silent electrical discharge and concentrations determined by μ C ozone sensors attached to a multiple point recorder. Twelve to sixteen values were averaged at each exposure level. Control animals were exposed to identical air flows containing 21% oxygen. After exposure, the mice were killed, their lungs aseptically removed and measurements of radioisotope and bacterial concentrations were made. In order to establish the number of bacteria represented by ³²P radioactivity, an Anderson sampler was attached to the aerosol chamber during the aerosol exposure period. Quantitative measurements of radioactivity and bacteria were performed from the 1.0–2.0 μ m sampling plate. A ratio of bacterial concentration to ³²P concentration was computed and designated the aerosol labelling ratio (*K*). The mathematical expression of pulmonary bactericidal activity for an individual mouse could then be computed from the formula in Table 1. The analysis of variance method was used to test for significance of difference.

With the exception of mice exposed to 4.25 p.p.m. of ozone, lung tissues were obtained from each group for histological examination. The data from the three aerosol runs are presented in Table 1. The range and mean values of ozone concentration are shown for each group. The lowest level of exposure was 0.62 ± 0.06 p.p.m. of ozone and the highest 4.25 ± 0.20 p.p.m.

Increased numbers of bacteria were consistently cultured from the lungs of animals exposed to ozone when compared with controls. The magnitude of the increases in bacterial numbers correlated with increases in ozone exposure levels up to 2.58 p.p.m. Mice exposed to 4.25 p.p.m. of ozone had similar numbers of staphylococci cultured from their lungs. Many of the mice exposed to 2.2 p.p.m. of ozone, or greater, yielded more staphylococci than were inhaled initially (negative bactericidal activity); apparently, intrapulmonary bacterial multiplication had occurred.

Comparison of the radioisotope counts for the 0 h control mice and mice sacrificed 5 h later demonstrated a 5–20% loss of radiophosphorus for each experiment. This loss of radioisotope is believed to be due to physical removal by the mucociliary stream and vascular absorption of detached isotope⁶. In two of the three aerosol experiments^{1,2} lower pulmonary radioisotope counts were observed in the ozone treated mice than in the controls. With the exception of the comparison of control mice and mice treated with 0.62 p.p.m. of ozone in the first aerosol experiments these differences were not significant ($P < 0.05$). The cause of the significantly lower radiophosphorus counts in the 0.62 p.p.m. group is not known.

Pulmonary bactericidal activity decreased progressively with exposure to increasing levels of ozone. In the individual aerosol experiments these differences were significant ($P < 0.05$) for the comparison of controls and mice treated with 1.10 p.p.m. of ozone or greater. The data from the three experiments were pooled and overall mean exposures and bactericidal activities were determined. According to these pooled data, significant differences in bactericidal activity between controls and treated mice occurred at each exposure level ($P < 0.05$).

Histological examinations of lung tissue showed a generalized dilation of capillaries and small vessels. These changes were more pronounced in mice exposed to the higher concentrations of ozone. Oedema was occasionally present within a few of the pulmonary alveoli of animals exposed to the higher ozone concentrations.

Two mechanisms of host defence against inhaled bacteria are known: mechanical removal in the mucociliary stream and destruction by pulmonary defence systems *in situ*. The ozone treated animals were able to remove similar numbers of radio-labelled bacteria to controls, presumably by the mucociliary stream, so the defect must involve intrapulmonary bacterial killing. Although there may be several bactericidal

Table 1 Effect of Exposure to Different Concentrations of Ozone on the Bacterial Activity of the Murine Lung

Set No.	Experimental group	Range	Ozone Mean	s.d.	Bacterial count in lungs *	³² P count in lungs †	Bacterial activity (%) ‡ **
1	Control 0 h (10)§					5,442 ± 301	
	Control 5 h (10)				168 ± 45	5,243 ± 476	84.7 ± 3.2
	Ozone (10)	0.53–0.75		0.62 ± 0.06	202 ± 44	3,868 ± 221	75.6 ± 4.7
	Ozone (10)	0.74–0.80		0.80 ± 0.03	508 ± 215	4,764 ± 385	49.6 ± 21.8¶
	Ozone (5)	2.04–2.40		2.23 ± 0.12	2,349 ± 952	4,116 ± 574	–178 ± 117
2	Control 0 h (10)					43,368 ± 3,801	
	Control 5 h (10)				260 ± 62	40,047 ± 3,391	85.9 ± 3.2
	Ozone (10)	0.53–0.88		0.70 ± 0.10	382 ± 93	36,134 ± 3,209	74.5 ± 6.1
	Ozone (10)	0.88–1.26		1.10 ± 0.09	833 ± 251	38,353 ± 6,212	51.3 ± 10.9¶
	Ozone (5)	2.50–2.86		2.65 ± 0.12	1,785 ± 251	37,583 ± 5,491	–3.4 ± 14.9¶
3	Control 0 h (10)					14,927 ± 1,285	
	Control 5 h (9)				220 ± 27	14,340 ± 893	84.4 ± 1.8
	Ozone (10)	2.02–2.58		2.28 ± 0.15	2,741 ± 445	14,437 ± 837	–45.4 ± 16.7¶
	Ozone (10)	3.85–4.48		4.25 ± 0.20	2,657 ± 389	13,282 ± 742	–102.7 ± 26.8¶
Overall mean ozone exposure and bacterial activity							
	Control (29)						85.0 ± 1.5¶
	Ozone (20)			0.66 ± 0.08			75.7 ± 3.8¶
	Ozone (20)			0.95 ± 0.16			50.3 ± 11.9¶
	Ozone (20)			2.39 ± 0.25			–99.4 ± 33.5¶
	Ozone (10)			4.25 ± 0.20			–102.7 ± 26.8¶

* Mean ± s.e. × 10².

† Mean ± s.e.

‡ Bacterial activity ± s.e.

§ Number in brackets is the number of animals in each group.

|| Mean ± s.d.

¶ *P* < 0.05.** Percentage bacterial activity = $\left[1 - \frac{\text{Bacterial count}}{^{32}\text{P}_+ (K)}\right] 100$.Bacterial count = bacterial count at 5 h; ³²P₊ = ³²P count at 5 h; and *K* = aerosol labelling ratio. *K* values for the three sets were: set 1 = 21.0; set 2 = 4.8; set 3 = 9.8.

mechanisms responsible for maintaining pulmonary sterility, previous investigations suggest that the macrophage performs the principal role⁷. Similar studies have also shown that conditions which depress macrophage function, such as hypoxia⁸, azotaemia⁹ or metabolic acidosis¹⁰, are associated with decreased pulmonary bactericidal activity. It is likely, therefore, that the inhibition in bactericidal activity we found is consequent on the toxic effect of ozone on the alveolar macrophage. This conclusion is consistent with data reported by Coffin and co-workers showing a progressive impairment of phagocytic function in pulmonary macrophages from mice exposed for 3 h to concentrations of ozone between 0.3 and 4.0 p.p.m.

Our findings may have clinical relevance. Exposures of this magnitude occur in various urban settings afflicted with smog. Although it is always hazardous to extrapolate from data obtained in laboratory models of infection to instances of human disease, nevertheless, the results of these investigations suggest that pulmonary resistance to infection may be diminished at exposure levels that are encountered on occasion in many cities. As a result, the human populations involved may be subjected to increased risk of respiratory infection.

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Enhancement of Residual Antibody Synthesis by Irradiation of the Spleen in Pre-immunized Rats

STIMULATION of the immune response by localized irradiation has been observed by several investigators^{1–3}. In all these studies, however, radiation was administered soon after antigen injection. In these experiments we have therefore examined whether localized irradiation of a lymphoid organ some time after antigen injection has a similar effect on residual antibody production. The effects of irradiation of the exposed spleen (with body shielding) on residual titres were studied in rats irradiated several weeks after a priming injection of sheep red blood cells (SRBC).

Female random bred rats weighing 200 g were used. Pre-immunization was effected with 1 ml. of a 1% SRBC suspension. Methods of irradiation of the exposed spleen with body shielding were as already reported⁴. For antibody titration rats were bled three days before radiation, thereafter each day during the first week and every other day for the two following weeks.

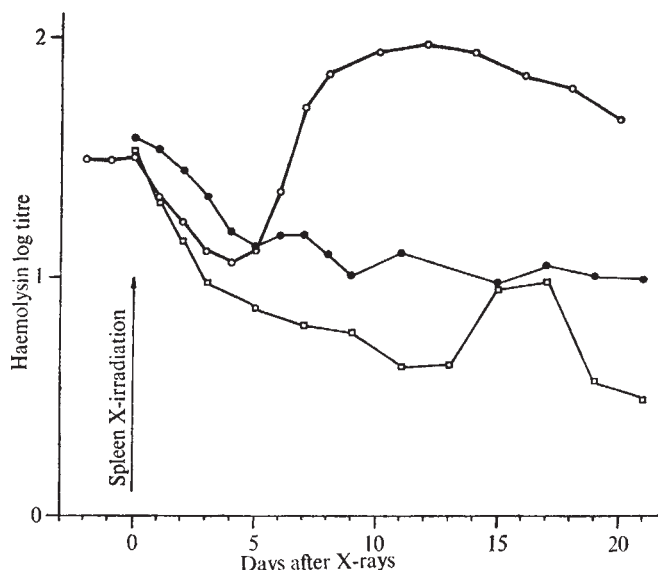


Fig. 1 Post-radiation recovery and overshoot of residual haemolysin titres in rats with spleens irradiated 5 weeks after the priming injection of SRBC (O—O). Note that the effect is absent when spleen irradiation is combined with splenectomy (●—●) or total body irradiation (□—□).

In the first experiment, 24 rats were injected with SRBC 5 weeks before irradiation of their exposed spleens with a heavy dose (1,000 or 10,000 rad of X-rays). We have found that pre-irradiation titres (mean 1.51 ± 0.04) decreased following the exposure (minimum 0.96 ± 0.10) but recovered in a few days, and then continued to increase, reaching levels which significantly exceeded the pre-irradiation titres (maximum 2.22 ± 0.7) (Fig. 1). The decline was similar to that seen in passively immunized rats indicating that radiation had temporarily suppressed the processes by which antibody levels are maintained. A mean titre of 0.96 ± 0.10 log units was found at maximal depression—a value which was markedly lower than the mean pre-irradiation titre. Occasional animals were observed with no detectable antibody but they all had low titres at the time of irradiation. Antibody began to increase again between days 4–6 post-radiation, and recovery, as gauged by a return to pre-radiation level, occurred in about 2 days. Thereafter titres continued to rise for 4–5 days. A mean titre of 2.11 ± 0.07 log units was reached at the post-radiation peak; this value was significantly higher than the mean pre-radiation titre ($P < 0.01$). In animals given 10,000 rad the overshoot phenomenon was, however, somewhat less marked.

The origin of recovery of residual titres was examined in a second experiment in which two groups of twelve rats each were primed with SRBC 5 weeks before irradiation of their spleens with 10,000 rad. Rats in the first group were splenectomized 4 days after spleen irradiation (at the beginning of the post-radiation antibody rise), whereas those in the second group were total body irradiated with 600 rad within 30 min after irradiation of their spleens, to destroy the circulating lymphocytes. In splenectomized animals titres declined for 4–5 days after spleen irradiation as in non-splenectomized ones, but thereafter they remained low and no return to pre-radiation level was observed (Fig. 1). Similar lack of recovery of titres was seen also in total body irradiated rats (Fig. 1). These are our conclusions: First, the spleen is the main site of production of both the residual pre- and post-radiation titres. Second, recovery of residual titres depended upon availability of radio-sensitive responsive cells from outside the spleen.

The cellular basis of the described phenomenon was studied in the third experiment in which 10,000 rad were administered to the spleen of two groups of rats: those injected with SRBC 5 weeks before the exposure and unimmunized controls.

Histological examinations of the spleen in these rats 2–10 days after spleen irradiation revealed that recovery resulted from immigration of circulating lymphocytes. These mobile cells endocolonize the follicular remnants which survive radiation and give rise to proliferating pyroninophilic cells. At the time when recovered residual titres have reached the post-radiation peak, numerous pyroninophilic cells were present in the red pulp together with a few small germinal centres in the reconstituted follicles. On the basis of the apparent similarity between these findings and those described by us earlier⁵ in rats injected with SRBC just before irradiation of their spleens, it seems reasonable to conclude that recovery and overshoot of residual titres represent a small secondary response which occurs without additionally injected antigen. Basically similar cellular changes were observed in both pre-immunized and unimmunized spleens; it seemed to us, however, that the overall cellularity of the red pulp was somewhat poorer in the latter.

Although the spleen was the main site of production of residual titres, and although the radiation doses were large enough to kill all lymphoid cells in the spleen, neither 1,000 nor 10,000 rad of X-rays were able totally to suppress its capacity for residual antibody synthesis. On the contrary, not only did residual titres recover within a few days but there was a marked overshoot. From parallel histologic studies it became clear that this was due to immigration of circulating lymphocytes which are able to repopulate the depleted spleen and generate new antibody-forming cells without further injection of antigen. At least two mechanisms may account for this. On one side, it is possible that the renewal of residual antibody production at a higher level represents a true *de novo* initiation of the immune response resulting from stimulation of repopulating immunocompetent cells by the persisting antigen. On the other hand, recovery and overshoot of residual titres may result from enhanced proliferation (because of a change in homeostatic regulatory mechanisms) of a few pre-existing antibody-forming cells which repopulate the spleen and are able to generate a synthetically active progeny without additional antigenic stimulation.

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Correlation between Psychotropic Potency of Psychotomimetic Methoxyamphetamines and their Inhibition of ³H-Normetanephrine Uptake in Rat Cerebral Cortex

THE psychotomimetic methoxyamphetamines are drugs which chemically resemble both mescaline and amphetamine. The subjective effects produced by them are similar to those elicited by other psychotomimetic agents^{1–3} and cross tolerance has

been demonstrated between at least one methoxyamphetamine and mescaline³. There may be some differences in the actions of these drugs and other psychotomimetic agents such as D-lysergic acid diethylamide (LSD), for in clinical trials⁴⁻⁷ and animal tests⁸ some methoxyamphetamines show considerable amphetamine-like activity, and one of them can produce marked enhancement of self-awareness in the absence of perceptual distortions and disorganization of thinking⁴⁻⁷.

The methoxyamphetamines are also valuable agents for psychotomimetic drug research because a large number of analogues have been synthesized and evaluated in human beings¹ and in animal tests⁹⁻¹¹. Of particular interest is the finding that small changes in chemical structure, particularly changes in the position of methoxy groupings, can produce drastic alterations in the psychotropic potency^{1,9}. For instance, in man, 2,4,5-trimethoxyamphetamine (2,4,5-TMA) is about twenty times as potent as mescaline, while 3,4,5-TMA is only twice as active as mescaline and 2,3,4-TMA seems to be devoid of psychotropic effect¹. These changes in the position of the methoxy groupings do not seem to cause any marked alteration in chemical properties such as their lipid solubility. It is also unlikely that these variations in psychotropic activity can be explained solely on the basis of differences in metabolic degradation, for the most and least psychoactive trimethoxyamphetamines are degraded by animals *in vivo* at similar rates (personal communication from C. Mitoma). It has been suggested that the psychotropic potency of these compounds is related to their tendency to assume conformations resembling certain of the ring structures of LSD^{12,13}.

Because of the variations in psychotropic activity of the different methoxyamphetamines, their differential effects on biochemical processes may provide a means of elucidating their mechanism of psychotropic action. Thus, if the capacity of these drugs to alter some biochemical process is closely correlated with their psychotropic potency, the alteration observed may be related to the mechanism of action of these agents. Because they are all derived from phenylethylamine, they might interact with norepinephrine neurones in the brain. The methoxyamphetamines are very weak inhibitors of the neuronal uptake of norepinephrine in the brain (ref. 14 and unpublished observations) and conceivably they might affect postsynaptic norepinephrine receptors.

There are no simple direct methods for biochemical studies of norepinephrine receptors. Recently, it was shown that the accumulation of norepinephrine by sympathetically innervated tissues extraneuronally (outside the sympathetic nerve ending) was antagonized efficiently by drugs which block norepinephrine receptors^{15,16}. This extraneuronal accumulation of norepinephrine occurs at low physiological concentrations of norepinephrine and seems to be similar to "uptake 2" of norepinephrine described by Iversen^{17,18}. In some tissues, it may be the principal mode of inactivation of the pharmacological actions of the amine¹⁹. Normetanephrine accumulates in sympathetically innervated tissues primarily by this extraneuronal mechanism²⁰. Because normetanephrine is taken up little, if at all, by sympathetic nerve endings, it could be a useful tool for studying the extraneuronal catecholamine uptake process. Recently, we found that normetanephrine is taken up by brain slices by a mechanism closely resembling the extraneuronal uptake of norepinephrine^{14,21}. Normetanephrine accumulation is unaffected by destruction of norepinephrine nerve terminals with 6-hydroxydopamine. Furthermore, drugs affect the uptake of normetanephrine and norepinephrine in the same way in rats pretreated with 6-hydroxydopamine. Adrenergic blocking agents are potent inhibitors of normetanephrine uptake in the brain¹⁴. We describe here a close correlation between the psychotropic potency of a series of methoxyamphetamines and their ability to inhibit the accumulation of normetanephrine by brain slices.

Male Sprague-Dawley rats (150-200 g) were killed by cervical fracture and their brains were rapidly removed and

chilled. The cerebral cortex was dissected, weighed and sliced to $0.1 \times 0.1 \times 0.5$ mm pieces with a tissue chopper²². Aliquots of brain mince (5 mg) were incubated with 2 ml. of modified Krebs-Henseleit medium¹⁴, nialamide (1.25×10^{-5} M), a monoamine oxidase inhibitor, and a gas phase of 95% oxygen and 5% carbon dioxide. Brain tissue was incubated with test drugs for 2 min after which ³H-normetanephrine (0.1 μ M, 3.8 Ci/mmole, New England Nuclear Corp.) was added and incubation was continued for 30 s. Incubation with ³H-normetanephrine was brief, because its accumulation is linear for only 60 s¹⁴. Incubation was stopped by pouring the tissue on Whatman No. 540 filter paper disks moistened with 0.9% NaCl in Gooch crucibles in a manifold vacuum assembly. After rinsing with 10 ml. of 0.9% NaCl, the filter disks containing tissue were extracted with 3 ml. absolute ethanol for 10 min; 10 ml. of toluene phosphor¹⁴ was added and the tritium content was assayed. In these conditions, ³H-normetanephrine accumulation was linear with time and with tissue concentration¹⁴. Normetanephrine accumulation was temperature dependent but unaffected by reserpine; it was saturable with a K_m of 1.4×10^{-3} M (ref. 14). Accumulated amine was localized in the soluble portion of sucrose homogenates. About 90-95% of accumulated tritium was unmetabolized ³H-normetanephrine¹⁴.

Table 1 Relation between Inhibition of ³H-Normetanephrine Uptake into Brain Slices by Psychotomimetic Methoxyamphetamines and their Psychotropic Potency

Drug	Inhibition of 30 s uptake of ³ H-normetanephrine	Psychotropic potency (mescaline units)		
		Man	Monkey ¹¹	Rat
2,4,5-TMA	7.20 $\pm$ 1.25 (4)*	17 ¹	5.4	7.2 ¹⁰ ; 4.0 ⁹
DOM	6.80 $\pm$ 1.07 (4)*	50 ³	—	25-50 ²⁶
2,4,6-TMA	5.00 $\pm$ 0.80 (3)*	10 ¹	3.2	1.7 ¹⁰ ; 1.0 ⁹
3,4,5-TMA	1.95 $\pm$ 0.35 (4)	2.2 ¹	1.8	4.0 ¹⁰ ; 1.5 ⁹
2,3,5-TMA	1.05 $\pm$ 0.35 (3)	4 ¹	1.1	—
2,3,4-TMA	0.78 $\pm$ 0.13 (4)†	< 2 ¹	0.7	1.6 ¹⁰ ; 0 ⁹

TMA refers to trimethoxyamphetamine; the preceding numbers refer to the location of methoxy substituents. DOM refers to 2,5-dimethoxy-4-methylamphetamine. Minces of rat cerebral cortex were incubated for 2 min with drugs after which ³H-normetanephrine (0.1 μ M) was added and incubation continued an additional 30 sec. ID-50 values were determined from log probit plots of percentage inhibition at three or four concentrations of inhibitor, in quadruplicate, after subtracting 0° C uptakes as blank. Psychotropic potency is expressed as mescaline units defined as the effective dose of mescaline divided by the effective dose of the compound tested. Effective doses in man are the minimally detectable dose¹ and for animals they are the ED-50 doses. Data were obtained from the references indicated. Relative potency was determined as the ratio of the K_m of dl-normetanephrine (1.4×10^{-3} M) to ID-50 of the test drug. Figures represent the mean $\pm$ S.E.M. and the number of separate determinations are in parentheses.

* These potencies do not differ significantly from each other but are all significantly more potent than 3,4,5-TMA ($P < 0.01$ for 2,4,5-TMA; $P < 0.01$ for DOM; $P < 0.025$ for 2,4,6-TMA).

† Significantly less potent than 3,4,5-TMA ($P < 0.025$).

The methoxyamphetamine compounds examined were active inhibitors of normetanephrine accumulation. There was a close correlation between their psychotropic potency in man and monkey (Table 1) and their ability to inhibit normetanephrine uptake. 2,4,5-TMA, 2,5-dimethoxy-4-methylamphetamine (DOM), and 2,4,6-TMA are the most active compounds in man and show similar potencies in their ability to inhibit normetanephrine accumulation. These compounds exhibited 5-7 times greater affinity for the normetanephrine transport system than did normetanephrine itself. By contrast, 3,4,5-TMA, 2,3,5-TMA and 2,3,4-TMA, which are weak or inactive agents in man, were considerably less potent inhibitors of normetanephrine uptake. 2,3,4-TMA, the weakest inhibitor of ³H-normetanephrine uptake, was slightly less active than 2,3,5-TMA and significantly less potent ($P < 0.025$) than 3,4,5-TMA. Although for some methoxyamphetamines there were species differences in psychotropic potency, 2,3,4-TMA was

invariably the least psychoactive methoxyamphetamine in all three species tested.

DOM has considerably greater psychotropic activity than 2,4,5-TMA or 2,4,6-TMA, while it is similar to them in its effects on normetanephrine uptake. DOM is an analogue of 2,4,5-TMA in which the methoxy at position 4 has been replaced by a methyl substituent. It was synthesized with the rationale of preventing possible metabolic degradation by demethylation of the methoxy group at C4 with subsequent excretion of the hydroxylated derivatives as water soluble conjugates². The possibility that its increased psychotropic potency is related primarily to metabolic degradation slower than the trimethoxyamphetamines rather than to enhanced potency at receptor sites is supported by the relatively prolonged action of DOM in "hippie" populations at high doses (personal communication from F. Meyers) and findings^{23,24} that 3,4-dimethoxyphenylethylamine is degraded primarily by demethylation at C4. The methyl group at C4 in DOM would prevent this metabolic route and could account for its increased potency.

Another interesting facet is that the relative potencies of the methoxyamphetamines in rats differ from their activities in man and in monkeys. Effects on normetanephrine uptake correlate best with psychotropic activity in man and monkey. These same species differences are also noted in the relative activities of the dimethoxyamphetamines in man and rat^{1,9,13}. It is conceivable that these species differences are related to variations in the metabolism of the aromatic ring of the amphetamines: Man and monkey metabolize the aromatic ring of amphetamine little if at all, while the predominant mode of metabolism of amphetamine in rats involves ring hydroxylation²⁵. The species differences in amphetamine metabolism may also apply to the metabolism of the methoxyamphetamines. Because metabolism of the aromatic ring of amphetamines is more extensive in the rat than in man and monkey, relative psychotropic potencies in these two species might reflect more closely the actions of these drugs at their receptor sites than do the *in vivo* potencies in rats.

Thus the correlation of the psychotropic activity of methoxyamphetamines with inhibition of normetanephrine uptake suggests that the sites of normetanephrine uptake may be related in some way to the locus of action of these drugs. A good candidate for such a site might be the postsynaptic receptors for norepinephrine in the brain.

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Stimulation of Intestinal Adenyl Cyclase by Cholera Toxin

THE molecular mechanism by which cholera toxin produces a massive loss of water and electrolytes is unknown, but it is clear that an increase in net secretion of approximately isotonic fluid occurs throughout the length of the small intestine^{1,2}. The relative contributions of increased active ion secretion and decreased active ion reabsorption to this fluid production are also unknown, although the fluid contains more HCO₃⁻ than does plasma^{1,3}. The toxin might stimulate secretory transport processes, for example Cl⁻ and HCO₃⁻, to such an extent that reabsorptive capacity is exceeded, water is held osmotically and massive diarrhoea results.

Recent reports suggest that increased concentrations of adenosine-3', 5'-monophosphate (cyclic AMP) in the intestinal mucosa are associated with the action of cholera toxin. Greenough *et al.*⁴ have shown that cyclic AMP, theophylline, two prostaglandins (PGA₂ and PGE₁) and cholera toxin stimulate fluid production in the ileum. Field *et al.*⁵ have reported that cyclic AMP and theophylline (which increases tissue concentrations of cyclic AMP by inhibition of phosphodiesterase) both stimulate short circuit current across the isolated rabbit ileum. Thus the action of cholera toxin to increase fluid production by the small intestine can be mimicked by agents which increase the intracellular concentration of cyclic AMP, and a change in electrolyte transport could be the responsible force. An increase in cyclic AMP in the cells can be achieved by either activation of adenyl cyclase, or by an inhibition of phosphodiesterase, and so we studied the action of cholera toxin on the activity of both these enzymes.

Preliminary experiments to investigate the effect of cholera toxin on the activity of phosphodiesterase and adenyl cyclase were carried out on six rabbits. Phosphodiesterase activity was determined by the method of Brooker *et al.*^{7,8} for measurement of cyclic 3', 5' AMP. In this method ³H-cyclic 3', 5' AMP is converted by tissue phosphodiesterase to ³H-5' AMP and the latter is converted to ³H-adenosine by 5' nucleotidase. The ³H-adenosine was separated from unreacted substrate with anion exchange resin and assayed by liquid scintillation counting. The assay procedure was carried out in a standard scintillation vial. Samples of the epithelial cell

homogenate (20 μ l.) were added to 30 μ l. of incubation mixture containing about 1 pmol of cyclic ^3H -AMP (about 15,000 c.p.m.), and incubated for 10 min at 30° C. The amount of protein (30–45 μ g) was chosen so that approximately 50% of the added ^3H -cyclic AMP would be destroyed during incubation. The final composition of the incubation mixture was as follows: ATP, 0.1 mM; Tris-HCl buffer, 20 mM (pH 7.5); ^3H -cyclic AMP, 20 μM ; MgCl_2 , 5 mM; and king cobra toxin, 0.4 mg/ml. The reaction was stopped by adding 1 ml. of AG1-X2 (200–400 mesh) resin in water (1:1). After subtracting the value of unquenched radioactive substrate (about 10% of the added c.p.m.) the results are expressed as pmol of cyclic AMP broken down by 1 mg of protein.

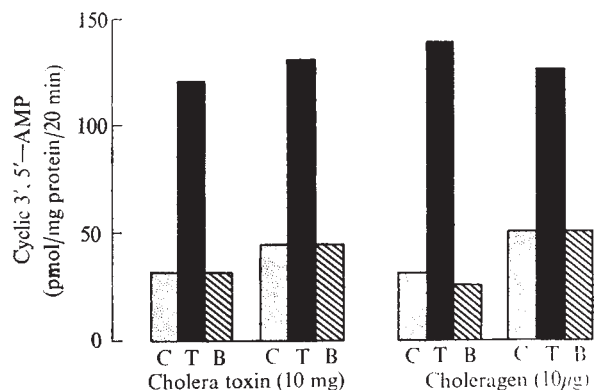


Fig. 1 Effects of cholera toxin and cholera toxin B on adenylyl cyclase activity in rabbit ileum. 10 mg of cholera toxin or 10 μ g of cholera toxin B was tested for activity on adenylyl cyclase, after 4 h incubation *in vivo*, and compared with the effects of boiled toxin and cholera toxin B. The results of individual experiments are shown. C, Control; T, toxin or cholera toxin; B, boiled toxin or cholera toxin B.

Crude exotoxin of *Vibrio cholerae* (Wyeth-NIH lot 001), supplied by Dr Qais Al-Awqati, was used as a freshly prepared solution. Cholera toxin (exo-enterotoxin elaborated by *V. cholerae* strain 561B Inaba) was a gift from Dr Richard A. Finkelstein and PGE₁ was a gift from Dr J. E. Pike of the Upjohn Co., Kalamazoo, Michigan. Adenosine-5'- α - ^{32}P -triphosphate (specific activity 4–6 Ci/mmol) was obtained from International Chemical and Nuclear Corporation, Irvine, California. ^3H -adenosine 3',5'-cyclic phosphate (specific activity 12.7 Ci/mol) was obtained from Schwarz BioResearch, Orangeburg, New York.

Ion exchange resin AG 50W-X2 (200–400, H⁺ form), AG1-X2 (200–400 mesh, Cl⁻ form), adenosine 5'-triphosphate disodium salt $-4\text{H}_2\text{O}$, adenosine $-3', 5'$ -cyclic phosphate 1/2 hydrate, 2'-phosphoenol-pyruvic acid-tricyclohexammonium salt, pyruvate kinase from rabbit muscle (300 IU/ml./9.3 mg protein) and myokinase from rabbit muscle (660 IU/mg protein) were obtained from Calbiochem, Los Angeles; snake venom from *Ophiophagus hannah* (king cobra) was obtained from Sigma Chemical Co., St Louis, Mo. All listed and unlisted chemicals were commercial preparations and were used without further purification. To prepare intestinal loops, laparotomy was performed on New Zealand white rabbits (2–2.5 kg) under intravenous sodium pentobarbital anaesthesia. After washing the succus entericus into a more distal portion of the intestine with 10 ml. of Tyrode solution, two or three loops, about 5 cm long, were prepared in each animal. Double sutures were placed between adjacent loops, and a rubber catheter (diameter 0.4 cm) was inserted to drain the loop. Care was taken that the blood supply to the loops was intact. Tyrode solution (1 ml.) with or without cholera toxin was injected into the loops and the abdominal wall closed.

To prepare tissue homogenate, intestinal loops were removed after incubation, separated from each other, and placed in ice cold Tyrode solution. Each loop was cut longitudinally and washed well in 15 mM Tris buffer, pH 7.5, with 12.5 mM MgCl_2 . The tissue was blotted on filter paper and the mucosal epithelial cells were scraped off with a glass slide. The cells were homogenized in a Ten Broeck ground glass homogenizer in 3–5 ml. of a 75 mM Tris-buffer solution with 12.5 mM MgCl_2 , pH 7.5. The protein content of the homogenate was estimated by the method of Lowry *et al.*⁶ and then diluted to give the required protein concentration for the enzyme assay. Bovine crystalline albumin served as the protein standard. In the experiments using a washed crude membrane fraction, the homogenate was repeatedly washed by suspension in 75 mM Tris-buffer with 5 mM MgCl_2 solution at pH 7.5 and centrifugation at 1,000g at +4° C. Adenylyl cyclase was assayed as described in the legend of Fig. 2.

Enzyme activities were estimated in the epithelial cells from adjacent loops of ileum, one of which had been treated intraluminally with 1 mg of cholera toxin for 4 h. The rate of breakdown of cyclic AMP by a homogenate from control tissue was 10.8 pmol/mg of protein/10 min and in that from toxin treated tissue 10.6 pmol/mg of protein/10 min ($\Delta=0.2\pm0.3$; $P=0.6$, $n=6$). Although no change could be detected in the activity of phosphodiesterase, there was a significant increase in the activity of adenylyl cyclase. The amount of cyclic AMP produced per mg of protein in 20 min was 46.9 pmol for control tissue and 65.8 pmol after toxin treatment ($\Delta=18.9\pm6.5$; $P<0.05$, $n=6$).

Because cholera toxin is sensitive to heat, it was important to test the effect of boiled toxin on adenylyl cyclase activity. Fig. 1 (left hand side) shows the results of individual experiments on the effect of cholera toxin and toxin which had been boiled for 10 min. After 4 h the activity of adenylyl cyclase in the homogenate from loops treated with cholera toxin was

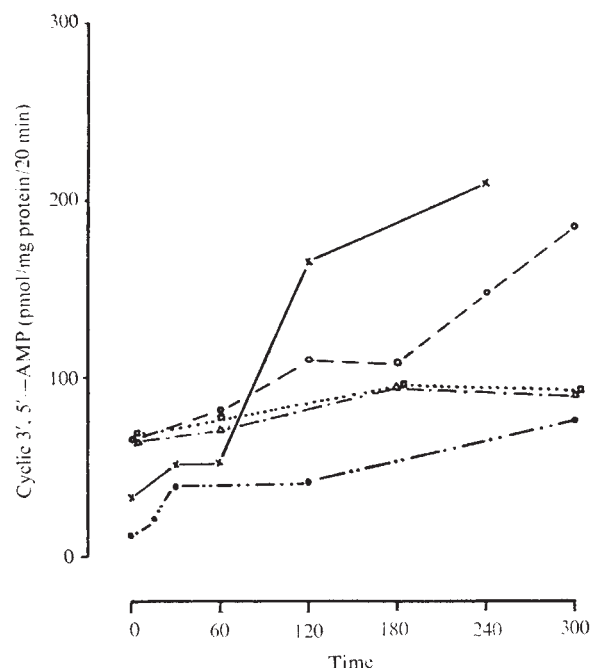


Fig. 2 Time course of activation of adenylyl cyclase by cholera toxin and cholera toxin B *in vivo*. The results of five individual experiments are shown. $\circ$, Δ , $\square$, 2 mg of cholera toxin per loop; $\times$, $\bullet$, 10 μ g cholera toxin per loop.

Adenylyl cyclase activity was determined by the method of Krishna *et al.*⁹. Samples of the epithelial cell homogenate of small intestinal mucosa (20–60 μ g of protein/20 μ l. of Tris-Mg buffer) were added to 30 μ l. of incubation mixture containing 1 μ l. of ATP- α - ^{32}P and incubated at 37° C for 20 min. The final composition of the incubation mixture was: Tris-HCl buffer, 30 mM (pH 7.5); cyclic AMP, 0.1 mM; ATP, 0.1 mM; MgCl_2 , 5 mM. Reaction blanks for each set of experiments were estimated in the absence of homogenate.

To terminate the reaction, 1 ml. of a solution containing 50 μ g of cyclic AMP, 100 μ g of ATP, and ^3H -cyclic AMP for calculation of recovery was added to each reaction mixture and immersed in boiling water for 5 min. The contents of the tubes were transferred to 0.6 $\times$ 4.0 cm chromatographic columns of AG 50 W-X2 (200–400 mesh) resin. The cyclic AMP was eluted with water in the fourth and fifth millilitre. Impurities in this fraction were removed by two precipitations with 0.25 M ZnSO_4 and 0.25 M Ba(OH)_2 , care being taken that the final pH of the solution was close to 7.5. After centrifugation for 10 min at 2,000g the precipitation was repeated. Then the supernatant was transferred to scintillation vials and ^3H and ^{32}P estimated by double isotope counting in a liquid scintillation counter. Recovery of cyclic AMP as estimated with ^3H -cyclic AMP was 50–70%.

The amount of cyclic AMP formed was calculated from the specific activity of ^{32}P -ATP in the incubation mixture and the amount of cyclic ^{32}P -AMP recovered minus reaction blank. Results are expressed in pmol of cyclic AMP/mg of protein/unit of time.

three to four times greater than that of the controls. The enzyme activity in the loops treated with boiled toxin, however, was equal to the control values. Similar results were obtained with a purified cholera toxin (Fig. 1, right hand side).

Fig. 2 shows the results of five time course studies carried out on individual rabbits. Different doses of toxin and cholera toxin were administered at particular times so that, at the end of the incubation, every intestinal loop could be removed and processed at the same time. Increasing activity of adenylyl cyclase was apparent throughout the time course for three of the animals. In two animals treated with a very small dose of toxin the increase occurred during the first 3 h.

Ileal loops in three rabbits were treated intraluminally with 10 µg of cholera toxin for 4 h. The homogenate from the epithelial cells was then centrifuged at 1,000g for 10 min and the supernatant was discarded. The procedure was repeated four more times and the washed crude membrane fraction was used for assay of adenylyl cyclase. Control loops were treated similarly. The enzyme activity in the washed membrane from cholera-treated loops relative to controls was increased by 120%, 265%, and 408% respectively in the three rabbits.

In four experiments the effect of a large dose of cholera toxin (100 mg per loop) was tested in rabbit jejunum. Incubation was for 4 h and large increases in adenylyl cyclase activity were produced by the toxin in all four rabbits (Table 1).

To investigate the interrelationship between two agents which stimulate adenylyl cyclase in intestine and the stimulation induced by cholera toxin, the effects of PGE₁ and sodium fluoride in cholera-treated and control tissue were examined. Preliminary tests on the concentration dependence of adenylyl cyclase stimulation by 10⁻⁸–10⁻⁴ M PGE₁ indicated that stimulation was maximal at 10⁻⁵ M. This concentration was therefore used for the studies.

When 1 mg of toxin/loop was used, there was a significant, 65%, increase in adenylyl cyclase activity. PGE₁ stimulated adenylyl cyclase by more than 60% in both control and toxin-treated loops. Furthermore, when the increments due to PGE₁ in control and treated preparations are compared (that is the activity in the presence of PGE₁ minus the basal activity) a significant enhancement of PGE₁ response is revealed in the tissue treated with cholera toxin. NaF markedly stimulated activity in both preparations, but no significant difference was detectable as a result of treatment with toxin (Table 1).

Table 1 Basal Activity of Adenylyl Cyclase and Activity stimulated by PGE₁ and Fluoride in Control Tissue and in Tissue stimulated by Cholera toxin and Cholera Toxin

Cholera toxin 1 mg/loop of ileum					
	Adenylyl cyclase activity				
	Control	Toxin	Δ + s.e.m.	P	n
Basal	54.7	90.0	35.3 ± 7.5	<0.001	18
PGE ₁	88.5	133.6	45.1 ± 8.9	<0.001	18
PGE ₁ -Basal	33.8	43.6	9.8 ± 3.6	<0.02	18
NaF	373.5	391.1	17.6 ± 22.6	0.50	18
NaF-Basal	318.8	301.1	-17.7 ± 21.8	0.40	18
Cholera toxin 100 mg/loop of jejunum					
	Control	Toxin	Δ + s.e.m.	P	n
Basal	43.4	148.6	105.2 ± 19.6	<0.02	4
PGE ₁	84.4	212.0	124.6 ± 25.1	<0.02	4
PGE ₁ -Basal	44.0	63.4	19.6 ± 5.7	<0.05	4
NaF	369.5	411.5	42.0 ± 44.7	0.40	4
NaF-Basal	326.1	262.9	-63.2 ± 52.4	0.30	4
Cholera toxin 10 µg/loop of ileum					
	Control	Toxin	Δ + s.e.m.	P	n
Basal	30.7	155.3	124.6 ± 27.2	<0.01	5
PGE ₁	62.7	185.8	120.1 ± 30.7	<0.02	5
PGE ₁ -Basal	32.0	27.7	-4.3 ± 10.0	0.70	5
NaF	337.4	331.9	-5.5 ± 31.9	0.90	5
NaF-Basal	304.4	176.5	-127.9 ± 27.9	<0.01	5

Incubation periods were 4 h.

The results obtained with jejunum and high doses of toxin were qualitatively similar to those obtained with ileum, but the magnitude of the change due to toxin was much greater, the enzyme activity being about 2.5 times that of the control preparation. The effect of PGE₁ was enhanced by cholera toxin and there was no significant difference in the fluoride response although the increase in activity due to fluoride was decreased (Table 1).

When 10 µg of cholera toxin was introduced into ileal loops, adenylyl cyclase activity was five times greater than in the control loops—the greatest activation we have seen in any series of experiments. The increase due to PGE₁ was unchanged while the increment due to fluoride was considerably decreased (176.5 pmol of cyclic AMP/mg protein/20 min compared with 304.4). The absolute level of activity reached by fluoride activation was the same in both control and cholera-treated tissue (337.4 and 331.9 pmol of cyclic AMP/mg protein/20 min respectively).

It has been known for many years, though not always accepted, that the massive loss of water and electrolytes in cholera patients occurs in the absence of histologically detectable damage to the intestinal mucosa^{10–12}. Evidence that damage to the mucosa is unlikely is available in reports of the low protein content of the cholera stool¹³ and in the demonstration that the rate of loss of macromolecules injected into the blood is unchanged by cholera¹⁴. It seems therefore that the most likely causes of fluid and electrolyte loss are: (1) increased secretion of electrolytes into the lumen of the intestine such that normal reabsorptive capacity is exceeded, or (2) that the active reabsorptive processes, chiefly sodium, are reduced.

Two recent reports become extremely relevant. Greenough *et al.* have demonstrated that cyclic AMP, theophylline, prostaglandins, and cholera toxin stimulate fluid production in rabbit ileum and that all four agents caused the production of fluid of similar composition⁴. Field *et al.* suggested that cyclic AMP and theophylline increase the secretion of chloride and probably also bicarbonate in rabbit ileum⁵. Therefore, an action of cholera toxin to increase the concentration of cyclic AMP in the intestinal mucosa could explain the electrolyte and fluid loss characteristic of the disease¹⁶.

We have shown that cholera toxin, when placed in the intestinal loop *in vivo*, increases the activity of adenylyl cyclase. The enzyme activity was increased to very high levels when large doses of toxin were used, in some cases as high as six times the control values. In experiments where adenylyl cyclase was significantly increased no effect on phosphodiesterase was detected. The toxin is sensitive to heat¹⁷ and boiled toxin is without effect on adenylyl cyclase activity (Fig. 1). Increased adenylyl cyclase activity has been demonstrated in both the lower ileum and the jejunum, a finding which is in agreement with an effect of this toxin on the whole of the small intestine. Increased enzyme activity has been detected as soon as 15 min after treatment with toxin, followed by a progressive increase in activity. This compares with a time course study in which there was only a slight delay, perhaps less than 15 min, between the application of the toxin and the stimulation of fluid production¹⁸. There was a subsequent steady increase in fluid production. It is possible, therefore, that the stimulation of adenylyl cyclase which we have observed is the mechanism by which cholera toxin produces its effects.

The molecular interaction by which cholera toxin increases the activity of adenylyl cyclase is uncertain. However, some characteristics of its action are helpful in understanding the mechanism. In the studies reported here the principal effect of cholera toxin is a large increase of basal enzyme activity without any change in the level of fluoride activation (Table 1). This strongly suggests that no synthesis of adenylyl cyclase occurs in toxin-treated tissue because fluoride activation should increase if the quantity of enzyme is increased. It is unlikely that an activation factor is released into the cytoplasm, or that cholera toxin is itself a soluble adenylyl cyclase¹⁸, because the increased enzyme activity after cholera toxin can

be observed in well washed membrane preparations. The latter suggests a relatively permanent change in the enzyme and may be due to the binding of toxin, or a fragment of toxin, by the enzyme. The interaction then results in both enhancement and modification of enzyme activity. For example, responsiveness to PGE₁ is increased by low doses of toxin but is not apparent when high doses are used. On the other hand, responsiveness to fluoride is decreased markedly by high doses of toxin. This (Table 1), which presents the results of cholera toxin treatment, may indicate the manner by which the toxin activates the enzyme. It suggests that toxin and fluoride stimulation are similar and that the toxin binds to the fluoride-sensitive site. Once bound it increases enzyme activity and diminishes additional stimulation by fluoride. Thus the effects of fluoride and cholera toxin are not additive but may be overlapping. Characterization of the interaction between toxin and enzyme is in progress.

In summary, the application of cholera toxin to the lumen of the small intestine results in a relatively permanent activation of adenylyl cyclase in the mucosal epithelial cells. There is indirect evidence that the increased activity of this enzyme stimulates fluid production in the intestine and that this may be due to stimulation of active anion transport processes, and so the effects of cholera toxin may be produced by the stimulation of adenylyl cyclase.

As the action of the toxin may result in acceleration of normal ion transport processes, we would like to draw attention to the following remarks on the action of cholera toxin, made in 1855 by Dr John Snow²¹.

"It would seem that the cholera poison, when reproduced in sufficient quantity, acts as an irritant on the surface of the stomach and intestine, or, what is still more probable, it withdraws fluid from the blood circulating in the capillaries, by a power analogous to that by which the epithelial cells of the various organs abstract the different secretion in the healthy body".

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Growth Potential of Scrapie Mouse Brain *in vitro*

STUDIES of mouse and sheep brain in tissue culture have shown that infection with scrapie increases growth potential¹⁻³. Glial cells always seem to predominate in these cultures, and in one case¹ there was a more rapid liquefaction of the ambient chicken plasma clot when explants of mouse brain infected with scrapie were used. Furthermore, the scrapie agent continued to multiply in a cell line which had been established from the brain of a mouse with clinical scrapie⁴. We have tried to quantify the growth potential of brain cells in culture by measuring their capacity to incorporate tritiated thymidine.

Small fragments of normal and scrapie-infected adult mouse brain of similar age were seeded on pairs of glass coverslips in 6 cm "escoplastic" Petri dishes, left for 1 h and then covered with 5 ml. of medium 199 (ref. 5) containing 20% foetal bovine serum. Cultures were incubated at 36° C in an atmosphere humidified with CO₂ and the medium was changed weekly, the amount of serum being reduced to 10% when cell growth had been established. Microscopic examination confirmed previous results: in one experiment infected brain grew well in seven out of eight dishes after 30 days, whereas growth was significantly poorer in three out of eight dishes seeded with normal brain, while in the other five there was no growth.

Just before it was used to measure the incorporation of ³H-thymidine, the medium was drained from each plate and replaced with 4 ml. of medium 199 containing 10% foetal bovine serum. Cultures were then pulsed with ³H-thymidine 2 Ci/mmol in a concentration of 2.5 µCi/ml. for various times. Cells were removed from the coverslips with 0.25% trypsin, counted and washed twice with Hanks basal salts solution. Their DNA, RNA and protein were precipitated with 5% trichloroacetic acid (TCA) after the addition of carrier protein. Precipitates were washed with TCA and absolute ethanol; they were dissolved in hyamine and mixed with a toluene based scintillation solution for counting.

Table 1 shows the results of preliminary experiments with labelling pulses lasting 5-70 h. In all cases infected cultures incorporated much more ³H, especially when pulsed for the longest time. Thus cultured adult brain which is infected with scrapie incorporates ³H from ³H-thymidine much more rapidly than does normal adult brain; the difference is of the same order as that between normal and "transformed" cells (our unpublished observations). Tritium is incorporated into TCA-insoluble material, not necessarily into DNA, because precursor may be degraded⁶ during the long pulse periods and some label may spread into RNA and protein⁷.

These and previous results¹⁻³ suggest that not only do cultures from adult scrapie brain grow more readily than do those from normal adult brain, but also that the individual cells are more active. In conjunction with the findings of Clarke and Haig⁴, our results do not support Adams's⁸ opinion "that there is at present little concrete evidence that infection with scrapie agent is associated with any specific effects on the growth potential of infected cells, or tissues". Much attention

Table 1 Tritium Incorporation into Normal and Scrapie Mouse Brain in Tissue Culture

Tissue	Period of growth (days)	Growth	³ H-thymidine pulse (h)	³ H incorporated (c.p.m./1,000 cells)
Adult normal mouse brain	30	One area of growth	70	19
Adult scrapie mouse brain	30	Confluent	70	732
Adult normal mouse brain	74	Several good areas	17	18
Adult scrapie mouse brain	34	Confluent	17	70
Adult normal mouse brain	75	One good area of growth	75	1
Adult scrapie mouse brain	35	Confluent	5	27
Primary rhesus monkey kidney *	9	Confluent	18	83
LLC-MK ₂ †	3	Confluent	18	596

* MRC Immunological Products Division.

† Passage 313 obtained at passage 292 from Flow Laboratories.

has been focused on the possible pathogenetic relationship between scrapie and multiple sclerosis⁹ and studies are in progress on labelled precursor incorporation into cultures of biopsy material from cases of multiple sclerosis and other neurological diseases as well as mouse scrapie.

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Functional Studies of Regenerated Muscles from Normal and Dystrophic Mice

MINCED skeletal muscle regenerates with autotransplantation and homotransplantation in the rat^{1,2}, and with heterotransplantation in the mouse³. If the distal stump of peripheral nerve is positioned in the proximity of the mince, motor units are believed to develop within 75 days, and stimuli applied to the nerve should elicit a contraction in the regenerate muscle (personal communication from Dr B. M. Carlson).

For homotransplantation of minced anterior tibialis muscle we used the 129/ReJ (Jackson Memorial Laboratory, Bar Harbor, Maine) normal mouse (+/+). Dystrophic mice (dy/dy) derived from this strain served for heterotransplantation of

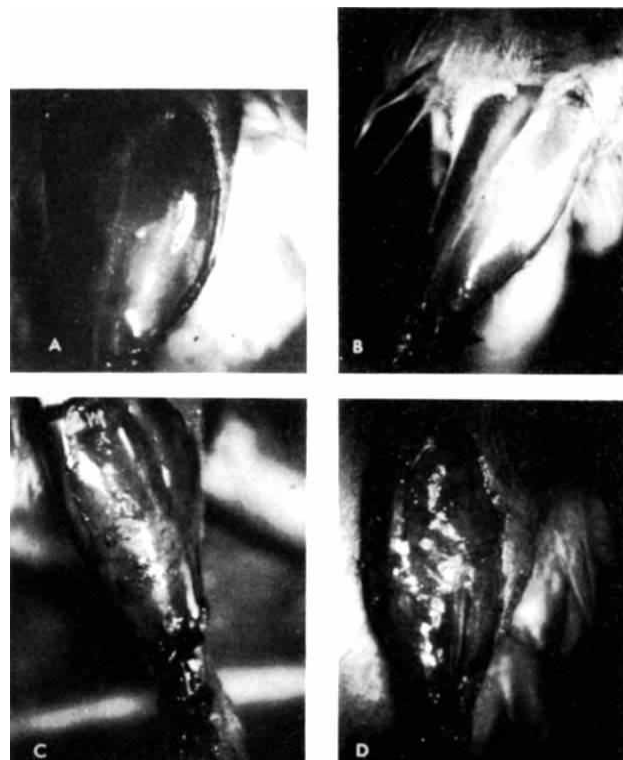


Fig. 1 A, Normal anterior tibialis; B, dystrophic anterior tibialis; C, normal regenerate from normal mouse; D, dystrophic regenerate from normal mouse (all $\times 40$). Normal and dystrophic muscles were 13–14 weeks old. All transplantations were carried out on anaesthetized (with 30 mg/kg sodium pentobarbitone) male mice 7 weeks old. In aseptic conditions whole anterior tibialis muscles were removed along with several millimetres of tendon, the nerve having been cut at its insertion into the muscle. Muscles were minced with a scalpel blade into fragments approximately 1 mm in size, then carefully laid into the existing muscle bed made available by resection of the counterpart muscle in the second mouse. Normal to normal transplantation provided approximately equal amounts of muscle mince to each mouse, but when dystrophic muscle mince was used about only half of the normal amount of tissue was available. This deficit was not corrected by adding more dystrophic muscle mince. The peroneal nerve was left in the vicinity of the mince. Transplanted mince soon formed a coagulum so that it was not necessary, in any way, to restrict the mince to the muscle bed before suturing the skin wound.

normal mince into dystrophic mice and of dystrophic mince into normal mice (for transplantation technique see legend to Fig. 1). The functional status of all these regenerates, as well as of control muscles, was determined. The complete experimental design is given in Table 1.

Homotransplanted normal mince regenerated by the seventy-fifth day. The regenerated muscles achieved normal twitch characteristics and gave rise to approximately 75% of control tension when indirectly stimulated within 75 days. Dystrophic mince transplanted into normal mice regenerated muscles with normal twitch characteristics, capable of developing tension approximately equivalent to normal regenerate muscle. Normal mince transplanted into dystrophic mice achieved no functional regeneration.

Fig. 1 depicts normal, dystrophic and regenerate muscles and Fig. 2 shows isometric twitch recordings from normal, dystrophic and regenerate muscles. Table 1 lists the results from all physiological measurements. Unoperated normal mice differed significantly (at or below the 5% level) from unoperated dystrophic mice in all parameters except contraction time (CT). The prolongation of the half relaxation time ($1/2RT$) and full relaxation time (full RT) is not as marked as reported by Sandow and Brust⁵ (129 strain), but their measurements were made in different conditions. Dystrophic muscles could develop only a quarter of peak tension, as compared with normals,

Table 1 Data from Muscles

	No. of mice No. of muscles	Geno- type of mouse	CT (ms)	1/2RT (ms)	Full RT (ms)	Peak twitch tension (g)	Peak tetanus tension (g)	Twitch/ tetanus	Muscle weight (mg)	Body weight (g)
Normal unoperated 13-14 weeks old	(6) (9-12)	+/+	11.08 ±0.30	7.00 ±0.22	24.09 ±0.74	11.41 ±0.50	37.60 ±1.36	0.303	35.59 ±1.45	27.88 ±1.37
Dystrophic unoperated 13-14 weeks old	(7) (9-12)	dy/dy	11.50 ±0.43	11.10 ±0.75	36.67 ±2.60	2.97 ±0.59	10.42 ±1.90	0.285	15.19 ±1.27	15.23 ±1.97
Normal regenerate 75 days of regeneration	(10) (7-9)	+/+	10.31 ±0.65	6.56 ±0.22	26.31 ±2.19	8.84 ±2.24	29.11 ±5.50	0.304	20.87 ±2.95	28.93 ±1.10
Contralateral side	(10) (8-9)		11.81 ±0.23	7.06 ±0.43	27.38 ±2.00	19.80 ±1.27	63.98 ±4.73	0.309	37.36 ±2.16	
Dystrophic regenerate 75 days of regeneration	(6) (6)	+/+	8.50 ±0.55	5.85 ±0.38	26.67 ±3.13	4.10 ±1.10	13.80* ±2.70	0.297	22.72 ±3.49	27.98 ±1.71
Contralateral side	(6) (5-6)		10.80 ±0.25	7.60 ±0.89	29.80 ±1.98	14.28 ±1.59	48.72 ±5.74	0.293	41.63 ±5.60	
Normal regenerate 75 days of regeneration	(6)	dy/dy	non-functional regeneration						21.84 ±2.78	18.67 ±0.63
Contralateral side	(6)		9.83 ±0.61	12.08 ±1.87	36.83 ±2.83	2.90 ±0.22	13.68 ±2.18	0.212		
	(6)									

*Four muscles only.

Where means and $\pm$ s.e. are given.

although the muscle mass was about 50% of normal. The higher values of peak tensions obtained in the contralateral muscles of operated normal mice can be attributed to hypertrophy.

Comparison of normal regenerates and dystrophic regenerates in normal mice revealed no significant difference (at or below 5% level) in any parameter. Most strikingly, there was no evidence of prolonged relaxation times in the dystrophic regenerate. The prolonged 1/2RT and full RT is a major defect in fast twitch dystrophic muscle and can be correlated to the observed electron microscopic and enzymatic changes seen in fragmented sarcoplasmic reticulum of dystrophic muscle^{8,9}.

The twitch/tetanus ratios of normal, contralateral and regenerated muscles were approximately equivalent. Dystrophic muscles differed substantially from these. The difference in

derived from dystrophic mince and allowed to regenerate in normal mouse. Isometric recordings from all muscles were made *in situ* at $34.5 \pm 1.0^\circ \text{C}$ and included measurements of contraction time (CT), 1/2 relaxation time (1/2RT), full relaxation time, peak twitch tension, and peak tetanus tension. After anaesthesia (with 30 mg/kg of sodium pentobarbitone) much of the skin over the tibia and above the ankle was removed. The tendon to the anterior tibialis was isolated, tied with fine thread and cut. Care was taken so as not to include the tendon of the extensor digitorum longus muscle which lies just beside that of the anterior tibialis. All tendons inserting in the ankle were cut and the anterior tibialis muscle was freed-up about its lower end. The peroneal nerve was exposed, and the mouse was laid in a specially designed mineral oil pool within a plexiglas chamber through which circulated warm water, the thermistor probe being in the oil or on the leg of the mouse. With the mouse lying on its back and side, the leg under examination was pinned to a fixed, cork-embedded shelf and the thread from the tendon attached to the transducer. Signals from the transducer (FTA-100-1 myographic force transducer, Hewlett-Packard, Sanborn Division, displacement $\pm 0.010''$, frequency response 390 c/s) in connexion with the carrier pre-amplifier were led directly to an oscilloscope (Tektronix 565). All measurements, excepting tetanus tension (slave polygraph), were made from photographs obtained directly from oscillographic tracings while the muscle length was set at optimum as has been described previously for neonate kittens⁴. Square wave stimuli (lasting 0.2 ms), supramaximal, were applied to the peroneal nerve through a glass-shielded, bipolar tungsten electrode, and the frequency of tetanic stimulation was 100 Hz. The central connexion of the peroneal nerve was cut or crushed and care was taken to observe that the neighbouring muscles, especially the extensor digitorum longus, did not contract. This was possible when stimulation was effected near the insertion of the nerve into the anterior tibialis. In regenerate muscles any adhesions to the extensor digitorum longus were severed, although the growth of connective tissue did not pose the problems anticipated. Frequently, direct stimulation was also carried out but all measurements are made from muscles stimulated indirectly.

peak tensions developed by normal and dystrophic regenerates is not as great as that in unoperated controls and dystrophic mice. It may reflect improved tension output in the dystrophic regenerate. Of the six dystrophic mice receiving normal mince, only one developed a functional regenerate, and this yielded a just perceptible contraction. It is unlikely that minced dystrophic muscle, when transplanted into dystrophic mice, would

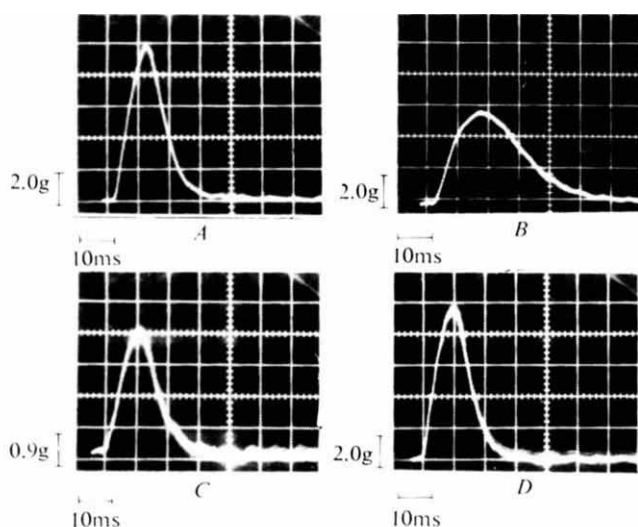


Fig. 2 Isometric twitch from (A) normal anterior tibialis in normal mouse; (B) dystrophic anterior tibialis in dystrophic mouse; (C) normal regenerate anterior tibialis derived from normal mouse mince; (D) dystrophic regenerate anterior tibialis,

also regenerate because dystrophic muscle cannot sustain a regenerative response⁶ and when injured it develops vacuolation, nuclear degeneration, and other deteriorative changes⁷.

McComas *et al.*¹⁰ have provided evidence in human muscular dystrophy for a major and possibly primary abnormality of the nerve supply to dystrophic muscles. Because dystrophic muscle appears normal when transplanted to normal mice, we conclude that dystrophic mouse muscle is rendered defective by the environment of the dystrophic mouse, and that muscular dystrophy in the mouse is not a primary myopathy.

The requirement of the peripheral nerve in many regenerating systems throughout biology has been well documented¹¹. Considerable indirect evidence has been adduced¹²⁻¹⁶ in support of neurotrophic regulation of skeletal muscle. Cross union of appropriate peripheral nerves, for example, tends to reverse physiological¹⁴, biochemical¹³ and histochemical¹³ characteristics of fast and slow twitch muscle. Recent reports of the transport of centrifugal protein within axons from the central nervous system to muscle help to define further the way in which "trophic" substances may reach muscle¹⁷. The relationship between rate of flow in axons and functional status of muscle has been supported by the finding that in "wobbler" mice with defective motor nerves, axonal transport of injected radioactive leucine is abnormal¹⁸.

These and our own findings provide compelling evidence that certain muscular dystrophies have their origin within the nervous system, probably within the motoneurone. The genetic defect may well result in the synthesis and/or transport of defective "trophic" substances which, when they interact within muscle, produce the irreversible changes characteristic of muscular dystrophy.

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Autolytic Enzymes in Growth of Bacteria

MANY bacteria lyse when growth of the cultures ceases, irrespective of whether growth has stopped because of the exhaustion of the growth medium or the addition of toxic substances such as the penicillins to the cultures. Lysis is caused by the presence in the microorganisms of autolytic enzymes (autolysins) which specifically hydrolyse mucopeptide polymers in the bacterial cell wall. The attack occurs, at least in some species, in a very restricted area around the point at which the bacteria will divide^{1,2}. This has led to suggestions that the autolysins are involved in bacterial growth. Three sorts of hypotheses have been suggested. First, that the enzymes are essential to make openings in the supposedly continuous network of mucopeptides in the wall^{3,4} so that the new building blocks can be added during growth^{3,5}; second, that the enzymes are essential for the remodelling of the wall in making the septum that divides one bacterium into two daughter cells^{6,7}; and third, that they are necessary to break the two newly formed bacteria apart from each other⁸.

The evidence supporting these hypotheses is indirect, with the possible exception of work with an amidase-negative mutant of pneumococci which can grow at a normal rate^{9,10}; moreover, some bacteria have more than one type of lytic enzyme. For example, bacilli often have two enzymes¹¹ which hydrolyse different bonds in their own mucopeptide. A possible, more direct way of investigating the role of these autolytic enzymes would be to obtain mutants either lacking the enzymes altogether⁴, or which have changed walls so that the enzymes can no longer hydrolyse them^{10,12}. If, of course, activity of the enzymes is essential for growth as it would be according to the first two hypotheses, then such mutants would have to be conditional (that is, the mutation must be only expressed in certain conditions and not others). We have isolated mutants of *Bacillus licheniformis* that are deficient in autolysin activity.

To isolate negative mutants, we designed a solid medium which contains preparations of the cell walls of the bacterial species under investigation, coupled with a red dye. Small amounts of cell wall can be used and zones of clearing around the wild type cells can be seen readily after suitable activation of the lytic enzymes whereas no zones are present around colonies of negative mutants. The cell wall preparation was made as follows. Cell walls were treated with sodium lauryl sulphate to inactivate their own lytic enzymes and with 0.1 M NH₄OH to remove ester alanine. Equal volumes of walls in suspension (10 mg/ml.), and 'Procion Brilliant Red' (ICI, Manchester) in a concentration of 160 mg/ml., both in 0.85% NaCl solution, were mixed. The mixture was stirred for 30 min at room temperature and an equal volume of 1.0 M NaHCO₃ buffer (pH 9.0) added. Stirring was continued for 90 min at room temperature, with a constant pH maintained by the addition of 2 M NaOH. The walls conjugated with the dye were then deposited by centrifuging, washed six times with water and dried from the frozen state.

When suspended in buffers such dye-conjugated walls from *B. licheniformis* were completely sedimented by centrifugation, leaving a clear supernatant solution. If, on the other hand, the conjugated walls were dissolved by either egg-white lysozyme or the lytic amidase from bacilli^{11,13}, 95% of the red dye was found in solution. When incorporated into agar growth media at the rate of 1 mg/ml., the medium was opaque and brilliant red; zones of clearing were produced by adding drops of lytic enzymes such as lysozyme or the amidase from bacilli but not by adding proteolytic enzymes such as trypsin or pronase. Heat-killed whole bacteria have been used in the past as an indicator of lytic enzyme production. Proteolytic enzymes, however, also dissolve heat-killed but not living bacteria, although they do not attack the cell walls.

When strains of *B. licheniformis* and *B. subtilis* were grown

Table 1 Some Properties of *Lyt*⁻ Mutants isolated from *Bacillus licheniformis* His⁻ Strain-6346

Micro-organism	Growth rate doubling time (min)	Rates of lysis of walls (wild type=100)			Analysis of walls (μmol/100 mg)			
		Endogenous autolysin	Enzyme added to inactivated walls	Amidase lysozyme	2,6-Diamino pimelic acid	Phosphorus	Uronic acid	Glucose
Wild type	84-96	100	100 (100)	100 (100)	34.9	103	36.4	30.6
<i>Lyt</i> ⁻ 3	83-103	2.5	— (7.5)	— (53)	57.0	172	2.5	3.0
<i>Lyt</i> ⁻ 4	180-200	6.0	10 —	57 —	41.0	145	35.0	9.0
<i>Lyt</i> ⁻ 5	99-104	1.1	1.9 (8.5)	76 (48)	64.0	170	2.6	3.1

All cell walls were prepared from cells grown on a minimal medium, except for those quoted in brackets which were prepared from cells grown in a casein hydrolysate medium. "Inactivated walls" refers to walls treated by sodium lauryl sulphate to inactivate their own lytic enzymes.

as colonies on such agars, only very limited zones of lysis were produced around them, but bacteriolysins could be demonstrated in the colonies by various means (for example, incubation of the agar plates in anaerobic conditions, treatment at 0°C or exposure to toluene vapour). After treatment of exponentially growing liquid cultures of *B. licheniformis* strain 6346 with a mutagen such as 1-methyl-3-nitro-1-nitrosoguanidine, mutant colonies without zones of lysis after an anaerobic treatment were found. Such isolates we intend to call *lyt*⁻. Mutants were also found which formed large zones of lysis even without treatment. These isolates we call provisionally *lyt*⁺.

Table 1 shows some properties of three representative strains of the *lyt*⁻ mutants obtained by the technique we have described. Some grew at about normal rates in minimal salts-glucose medium, but one, *lyt*⁻4, grew more slowly. Cell walls isolated from them lysed at from 1-6% of the rate of walls from the wild type. The very slow rate of lysis was caused by residual action of the amidase; and the walls were relatively resistant to externally added amidase but sensitive to lysozyme. Autolytic glycosidase activity seemed to be normal though it was very low both in the mutants and in the wild type. Two of the least lytic of the mutants (*lyt*⁻3 and 5) grew as very long chains of unseparated bacilli. The walls of these two mutants were different from the wild type in that they contained more mucopeptide as shown by their increased content of α-ε-diaminopimelic acid (Table 1), very little teichuronic acid^{14,15} (low uronic acid, see Table 1), slightly more phosphorus (P) presumably as glycerol teichoic acid; but they had a low content of glucose. *Lyt*⁻4 again differed from the others.

This work suggests that one role of the autolytic amidase in bacilli, which is the most prominent autolysin in this genus, is concerned with the separation of the individual bacteria. The failure of the walls to lyse in the mutants may result rather from changed wall chemistry, as has already been shown with pneumococci^{10,12}, than from loss of the lytic enzyme; but this problem is not yet unambiguously solved, for a reduction in the normal amount of amidase might lead to the abnormal wall chemistry. The growth rates of the two least lytic mutants were similar to that of the wild type. It is therefore possible to say that the autolytic activity of *B. licheniformis* can be grossly reduced without significant alteration of the growth rate of the bacteria.

It should be pointed out that no genetic evidence is yet available for these mutants and it is not possible to say whether their properties all arise as the result of mutation at a single point.

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R Factor with a Mutation in the Tetracycline Resistance Marker

THERE is evidence that resistance to tetracyclines (TCR) in Gram-negative bacteria that is mediated by R factors involves an inducible mechanism. Thus two levels of resistance to tetracyclines have been demonstrated in various strains of *Escherichia coli* harbouring R factors^{1,2}. R factor bearing cells cultured in the absence of tetracycline are about 50% resistant when challenged with 50 μg of tetracycline/ml., as can be seen from the effect of the drug on the level of protein synthesis in the cells². Cells cultured in the presence of a sub-inhibitory concentration of tetracycline, however are not significantly inhibited by a subsequent challenge with 50 μg of tetracycline/ml.; the 50% level of inhibition is only reached at concentrations in excess of 150 μg/ml. This adaptation, which is apparently associated with a decrease in the absorption of tetracycline by the cells, can be suppressed with inhibitors of protein and RNA synthesis². The tetracycline resistance mechanism thus resembles an inducible system although the end products of the resistance gene(s) have not yet been identified.

If the genetic components of the tetracycline resistance marker involve the usual components of an inducible system—repressor, operator and structural genes—it should be possible to obtain a "constitutive" mutant in which tetracycline resistance is always expressed at a high level because of complete or partial loss of repressor control.

We have isolated such a mutant after subjecting a culture of *E. coli* harbouring an R factor with a tetracycline-resistance marker to mutagenesis with N-methyl-N-nitroso-N'-nitroguanidine (200 μg/ml.). The cells were allowed to recover in a growth medium for 16 h after mutagenesis, followed by growth on 'Oxoid' membrane filters overlying nutrient agar plates. The filters were then transferred to MacConkey plates containing 125 μg of tetracycline/ml. and in which

the sole source of nitrogen was ammonium sulphate. Colonies which turned pink within 1.5 h were considered likely to be either *lac* constitutive mutants or TCR constitutive, for the wild-type colonies did not turn pink on these plates within this time unless induced to high-level tetracycline resistance by pre-exposure to a sub-inhibitory concentration of antibiotic.

Table 1 Resistance of R Factor-bearing Strains of *E. coli* to Tetracycline

Concentration of tetracycline in second incubation ($\mu\text{g/ml.}$)	Inhibition of protein synthesis by tetracycline in second incubation (%)			
	First incubation without tetracycline		First incubation with tetracycline	
	TCR	TCRM1	TCR	TCRM1
10	0	0	0	0
50	56	0	8	0
100	90	25	10	0
125	99	49	28	22

The resistance of the parent strain of *E. coli* harbouring a wild type R factor (TCR) and that of the mutant (TCRM1) was assayed by determining the inhibition of protein biosynthesis by tetracycline. Both strains were first preincubated at 37° C for 30 min in a complex medium² with or without 10 μg of tetracycline/ml. The cells were harvested by centrifugation and resuspended in a simple salts-glucose medium² containing 0.2 μCi of ¹⁴C-leucine/ml., 0.2 mM and the indicated concentration of tetracycline. After incubation at 37° C for 20 min the incorporation of ¹⁴C-leucine into cell protein was measured as described previously². In non-resistant strains of *E. coli* protein synthesis is completely inhibited by 10 μg of tetracycline/ml.

By this technique, a mutant strain of *E. coli* (TCRM1) was obtained which seems to be largely constitutive for tetracycline resistance. Table 1 shows the effect of various concentrations of tetracycline on the level of protein synthesis in both the R factor-bearing parent cells and in TCRM1. The mutant was markedly more resistant to tetracycline than the parent strain when neither strain had been pre-exposed to a sub-inhibitory concentration of drug (10 $\mu\text{g/ml.}$). At challenge concentrations of 100 and 125 μg of tetracycline/ml., however, there was an indication that pre-exposure of the mutant to a low concentration of tetracycline had further increased the resistance of these cells. Tetracycline resistance could be transferred from TCRM1 to a suitable recipient strain of *E. coli* in mating experiments with the same frequency that was observed for the parent strain (10⁻⁵). The resistance remained partially constitutive in the recipient organism.

The isolation of R factor-bearing *E. coli* with the modified type of tetracycline resistance described strengthens our belief that the resistance carried by the wild type R factor is an inducible system. In cells cultured in the absence of antibiotic, resistance is expressed at a low level presumably because of limited transcription and translation of the TCR structural genes. Treatment of the cells with minute quantities of tetracycline (as little as 0.05 $\mu\text{g/ml.}$) apparently derepresses the TCR genome making possible the expression of resistance at its highest level. The nature of the mutation in cells bearing the TCRM1 type R factor that results in extensive derepression of the resistance mechanism is not known. It may involve either the repressor or operator genes, for mutations in either of these could result in a partial loss of repressor control.

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Hydrocortisone Resistance of Activated Initiator Cells in Graft versus Host Reactions

ALTHOUGH corticosteroids are usually considered to be lympholytic and immunosuppressive, some immunocompetent cells, presumably lymphoid, resist the actions of corticosteroids; for example, the graft versus host cell in chicken¹ and mouse² thymus, and the bone marrow antibody-forming cell precursor³. We have shown⁴ that the cell(s) in mouse spleen which can initiate graft versus host (GVH) reactions are resistant to the action of hydrocortisone. In these experiments, hydrocortisone was given to normal mice of parental strains. Two days later the surviving spleen cells (about 20%) were transferred to adult F₁ hybrid recipients, where they showed unimpaired GVH responses as measured by splenomegaly. In these circumstances, the GVH initiator cell was exposed to hydrocortisone before coming into contact with antigen (mainly H2), and it was thus unlikely to be dividing rapidly, if at all⁵. Because there is evidence that corticosteroids have their greatest effects on rapidly dividing cells^{6,7} we have investigated the hydrocortisone sensitivity of GVH cells during antigenic stimulation, when thymus-derived cells are known to proliferate^{8,9}.

In a first series of experiments, normal parental spleen cells were injected into unirradiated adult F₁ mice. GVH was assessed by splenomegaly in the recipients. Hydrocortisone was given to the recipients at various times after cell transfer. Donors were 10 week old female BALB/c mice or, for controls, 8 week old CAF₁ mice (A × BALB/c); recipients were 8 week old male CAF₁. On day 0, four groups of six CAF₁ each were given 10 × 10⁶ BALB/c spleen cells intravenously (test set), and four similar groups were given 10 × 10⁶ CAF₁ spleen cells (control set). One test and one control group were untreated, one group of each set received 2.5 mg of hydrocortisone acetate suspension (Pfizer) intraperitoneally on day 0 ("early treatment"), one group of each set received 2.5 mg of hydrocortisone on day 6 ("late treatment") and the fourth pair of groups received 2.5 mg of hydrocortisone on days 0, 2, 4 and 6 ("treatment throughout"). All animals were killed on day 8, and the spleens were weighed (Table 1). The increase in spleen weight due to GVH is 65 mg (group A minus group E). Of this, approximately half can be suppressed if a single dose of hydrocortisone is given early (group B minus group F) or late (group C minus group G) in the course of the reaction. By comparing the two groups treated throughout (D and H) it may be seen that all the splenomegaly arising from the GVH reaction is suppressed in the constant presence of hydrocortisone.

Table 1 Spleen Weights of CAF₁ Mice given BALB/c or CAF₁ Spleen Cells on Day 0 and Hydrocortisone at Various Times

Group	Donor strain	Hydrocortisone	Spleen weight (day 8)
A	BALB/c	None	177 (171-184)
B		Day 0	96 (71-122)
C		Day 6	91 (67-115)
D		Days 0, 2, 4, 6	48 (40-56)
E	CAF ₁	None	112 (91-133)
F		Day 0	60 (52-69)
G		Day 6	54 (48-61)
H		Days 0, 2, 4, 6	40 (32-48)

All recipients received 10 × 10⁶ cells on day 0. Hydrocortisone acetate (2.5 mg) was given intraperitoneally at the times indicated. Spleen weights are in mg, with the 95% confidence limits in parentheses.

It is not possible to determine which cells are affected by hydrocortisone from this type of experiment alone, because it is well established that both donor and recipient cells multiply to produce splenomegaly in GVH reactions¹⁰. Consequently, we tested the effect of hydrocortisone on the GVH initiator in the absence of a host response. A two-stage transfer

system, based on the method of Cerottini *et al.*¹¹, was used. Parental spleen cells were immunized to the foreign H2 antigens by injection into lethally irradiated F₁ hosts; the spleen cells of these primary recipients were transferred after 5 days to normal F₁ hosts for assay of GVH. Fifty million BALB/c (test) or CAF₁ (control) spleen cells were transferred intravenously on day 0 to groups of CAF₁ recipients irradiated with 900 rad ⁶⁰Co. One test group and one control group were given 2.5 mg of hydrocortisone on days 1 and 3; another pair of groups was untreated. On day 5 the spleens were removed, cell suspensions made and the cells were transferred intravenously to normal CAF₁ so that each secondary recipient received the equivalent of one spleen. Spleen to body weight ratios in the secondary recipients were determined after 8 days as an index of GVH. (Significant immunization of the parental spleen cells had occurred, for earlier experiments had shown that no GVH ensued if immunization was allowed to proceed for 1 day.) This immunization was not significantly decreased by hydrocortisone (Table 2). There was no detectable carry-over of hydrocortisone from the primary to the secondary recipients (compare group C with group D).

Table 2 Effect of Hydrocortisone on the Sensitization of BALB/c Spleen Cells to CAF₁ Histocompatibility Antigens

Group	Donor (day 0)	Hydrocortisone on days 1 and 3	Cells/spleen of primary recipient	Spleen index of secondary recipient
A	BALB/c	No	19 × 10 ⁶	1.49 (1.26–1.73)
B		Yes	12 × 10 ⁶	1.36 (1.20–1.51)
C	CAF ₁	No	9 × 10 ⁶	1.00 (0.88–1.12)
D		Yes	7 × 10 ⁶	1.04 (0.95–1.13)

Primary recipients were CAF₁ irradiated with 900 rad and given 50 × 10⁶ BALB/c or CAF₁ spleen cells. Hydrocortisone was given intraperitoneally (2.5 mg/dose). On day 5, primary recipient spleens were removed, and cells equivalent to one spleen were transferred to normal CAF₁ secondary recipients. Eight days later, these animals were killed and spleen weight (mg) to body weight (g) ratios were calculated. Spleen index is the spleen to body weight ratio of the test group divided by the spleen to body weight ratio of the control group (group C); 95% confidence limits are in parentheses.

These two series of experiments together with our previous work⁴ indicate that the GVH initiator cell(s), presumably lymphoid, is resistant to hydrocortisone whether it is in the unstimulated state, or whether it is responding to histocompatibility antigens. Almost complete suppression of splenomegaly is obtained, however, when the recipient is treated with hydrocortisone throughout the GVH reaction. This suppression is considered to be caused by inhibition or killing of the recipient's GVH-responding cells, whose origins are unknown. Some GVH-responding cells, or their precursors, may survive hydrocortisone, for hydrocortisone given to recipients only once during the reaction suppressed it only partially.

These results suggest that corticosteroids would be poor drugs for the prevention or treatment of GVH reactions because they do not eliminate the cells which initiate the immunological attack. We are investigating the possibility that, in general, corticosteroids affect the visible manifestations of cell-mediated immunity by suppressing the non-specific response to specific immunological events.

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Selective Predation by Newts on Frog Tadpoles treated with DDT

HYPERACTIVITY induced by exposure to a pesticide could be a disadvantage to an organism in the wild^{1–5}, particularly by increasing the risk of predation. I have therefore investigated whether warty newts (*Triturus cristatus*) will preferentially prey on hyperactive, DDT-treated tadpoles of the common frog (*Rana temporaria*) rather than untreated tadpoles.

Each newt was kept in a glass tank 40 cm long × 20 cm × 20 cm, filled to a depth of 7.5 cm with tap water and with a floor covering of fine gravel. Once every 2 days, twelve live frog tadpoles (40–100 mg) were introduced into each tank as food. The normal hunting technique of these newts comprised (i) locating a tadpole, (ii) stalking it to within 2 cm, and (iii) making a snapping lunge which was usually unsuccessful. Males were used whenever possible because they were more voracious.

I have already found that *pp*'DDT induces a series of quite distinct behavioural changes in frog tadpoles. At first, tadpoles enter a "frantic" phase of tail lashing, body twisting and rapid swimming, followed by a "resigned" phase characterized by persistent slow swimming in a twisting manner. Finally, the animals become moribund and die. Frantic and resigned tadpoles are considerably hyperactive, and animals in both phases have been tested against controls for selective predation.

Tadpoles were chosen either without hind limbs or with small hind limb buds (younger than stage IV (ref. 6)). They were treated with *pp*'DDT in groups of forty as before⁵, except that insecticide was added to amphibian saline in ethanol using solutions from the organochlorine test kit for mosquito larvae supplied by the World Health Organization. After exposure, tadpoles were transferred to fresh saline and ten of them were removed, weighed and analysed for pesticide by gas-liquid chromatography using an electron capture detector (Table 1).

The remaining tadpoles were used in the trials within 2 h of exposure to DDT to ensure that their insecticide content did not decrease by more than about 5% (ref. 5). In each trial a treated tadpole and a control of similar size were introduced together into a tank at the opposite end from the newt. Lunges made by the newt were recorded until a tadpole was caught. The surviving tadpole was removed immediately and a fresh pair was introduced. For trials on each behavioural phase four newts were used. By testing a group of tadpoles on each of two successive days, it was intended that each newt should participate in thirteen trials, but during the frantic phase trials, one newt—the only female—caught only nine tadpoles in the two 2 h periods (Table 2).

The tendency to catch the treated tadpole of each pair was highly significant (frantic, $\chi^2 = 30.1$; resigned, $\chi^2 = 33.9$; $P < 0.001$ for both), only ten control tadpoles being taken out of a total of 100 trials. Significantly more initial lunges were made at the hyperactive tadpole of each pair (frantic, $\chi^2 = 21.3$;

resigned, $\chi^2=37.2$; $P<0.001$). These newts probably hunted primarily by sight, although sense of smell can be important⁷ and in special circumstances sense of touch may also be used⁸. Hyperactivity of the treated tadpole has apparently caused the newt to locate it more readily and has then stimulated an attempted kill. A laboratory study with a hawk and coloured mice has already suggested that conspicuousness rather than oddity controls the selection of prey⁹.

More hyperactive tadpoles have been killed because signifi-

Although this experiment indicates that hyperactivity caused by a pesticide will be an additional hazard to a tadpole in the wild, the effect on a population is not necessarily harmful. If poisoned tadpoles survive predation and their activity returns to normal, they might die after the metamorphic climax when small residues are apparently mobilized⁵. Increased predation of hyperactive tadpoles may therefore be beneficial to healthy individuals that have not been exposed to pesticide by preventing pointless competition.

Table 1 Treatment, Wet Weight and Pesticide Content of Tadpoles

	DDT in amphibian saline (p.p.m.)	Treatment time (h)	Group	*Mean wet weight of one tadpole (mg)	*Mean pesticide content of one tadpole (μ g)	
					<i>pp'</i> DDT	<i>pp'</i> DDE
"Frantic" phase tadpoles	0.05	5	1	82	0.50	<0.001
			2	63	0.61	<0.001
"Resigned" phase tadpoles	0.05	19	3	54	0.75	0.01
			4	50	1.20	<0.001

* Derived from a bulked sample of ten tadpoles from each group. In controls the mean *pp'*DDT and *pp'*DDE content <0.001 μ g/tadpole.

Table 2 Lunges made by Newts during Predation Trials

	No. of trials		No. of initial lunges	Total No. of lunges	No. successful	No. unsuccessful
Frantic phase trials	48	At hyperactive tadpoles	40	167	43	124
		At control tadpoles	8	38	5	33
Resigned phase trials	52	At hyperactive tadpoles	48	144	47	97
		At control tadpoles	4	15	5	10

Table 3 Mean Wet Weight and Pesticide Residues in Newts 1 Week after Trials $\pm$ s.e.

	Mean wet weight (g)	Mean pesticide content (p.p.m.)		<i>pp'</i> DDT % recovery
		<i>pp'</i> DDE	<i>pp'</i> DDT	
Control newts	4.8 $\pm$ 0.2	0.36 $\pm$ 0.14	<0.002	
Newts used in frantic phase trials	6.4 $\pm$ 0.8	0.23 $\pm$ 0.01	0.62 $\pm$ 0.04	65 $\pm$ 7
Newts used in resigned phase trials	4.9 $\pm$ 0.5	0.65 $\pm$ 0.11	1.38 $\pm$ 0.28	54 $\pm$ 5

cantly more lunges have been made at them (frantic, $\chi^2=81.2$; resigned, $\chi^2=105$; $P<0.001$), there being no significant difference in the percentage of successful lunges at treated and control tadpoles (frantic, $\chi^2=2.7$; resigned, $\chi^2=0.003$). The lack of response by treated tadpoles to external stimuli should make them easier to catch⁵, but this factor may have been counterbalanced by their persistent, uncoordinated movements.

Seven days after the end of the trials, each newt was weighed and analysed for organochlorine residues, and so were four untested controls obtained from the same source (Table 3). The *pp'*DDT intake of each newt can be estimated from the number of treated tadpoles consumed and their pesticide content. Analysis of the newt carcasses revealed that most of the *pp'*DDT could be accounted for (Table 3). The metabolite of *pp'*DDT most commonly found in vertebrate tissues, *pp'*DDE, was present in every sample, and one control newt contained the relatively high concentration of 0.76 p.p.m. This variability makes it difficult to determine the extent of metabolism. The higher concentrations of *pp'*DDE in newts used in the resigned phase trials suggest that metabolism occurred, although analysis of variance shows no significant difference.

The acquired pesticide residues had no visible effect on the newts, but exposure was limited to 2 days, and it is likely that they are more resistant than the tadpoles⁵. If selective predation can be extrapolated to other predator-prey systems, toxin-susceptible predators taking relatively resistant prey will be especially at risk.

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BOOK REVIEWS

Duality of Science

Beyond the Ivory Tower: The Frontier of Public and Private Science. By Sir Solly Zuckerman. Pp. ix + 243. (Weidenfeld and Nicolson: London, November 1970.) 50s.

SIR SOLLY ZUCKERMAN has produced out of his experience both as a mammalian anatomist and as Chief Scientific Adviser to the British Government an intriguing account of the two faces of science. The first face is that of "private science", the way in which a scientist makes a discovery, demolishes a hypothesis, attempts to convince his colleagues; the second face is that of "public science", the role of a scientist as an adviser, the application of science to social needs, the way in which decisions about the use of applied science for national defence or economic growth have to be taken. Sir Solly is a distinguished practitioner in both fields and he believes that the two activities have different criteria of truth. Pure science is only concerned with objective truth; with the hypothesis whose predictions can be either verified or shown to be false. On the other hand, applied science, technology, science in the area of political judgments, requires decisions which involve subjective values (should we devote money and manpower to building a supersonic transport or should we devote the same effort to improving communications between properly sited airports and the capital? Should we spend money on geriatric research or on heart transplants?).

Sir Solly is of course quite right to stress the objective nature of "pure science" and the fact that scientists are weak, fallible creatures with appallingly subjective responses should not obscure this. The separation of "public" and "private" science is, however, perhaps not quite so clear to many of us as Sir Solly suggests. Some branches of basic science are now so expensive in national terms that scientists cannot be solely responsible for decisions about them. They can only be responsible within an overall budgetary framework. For a country of limited financial resources, such as Great Britain, decisions about spending on "Big Science" and "Little Science", pure science though both may be, will be political decisions. Questions such as "Can a nation like Great Britain afford to support basic research in high energy nuclear physics?" have a subjective value judgment built into them.

Both parts of the book are controversial, the section on "Private Science" perhaps more so than Sir Solly realizes. This part consists of three stories, which illustrate what science is about and how it progresses, drawn from Sir Solly's own

specialism. The first shows how an accepted scientific hypothesis was proved to be incorrect, and how Sir Solly demonstrated that the mammalian ovary at birth has a finite stock of egg cells which decline in numbers. The account illustrates pretty clearly how difficult it is to dislodge a mistake which has become sufficiently firmly embedded in the minds of scientists and in the scientific literature.

The second story relates Sir Solly's failure to dislodge a set of hypotheses on the mechanism by which the brain controls the pituitary gland. I found this example, for a non-biologist, pretty difficult to follow and too technical to judge the relative merits of the two sets of views. The third concerns the determination of the status of *Australopithecus* from a comparison of the fossil skulls and other remains that we have with those of man and of the great apes. It confirms a view I had held, that comparative anatomy is more of an art than a science; related more closely perhaps to an art expert's identification of a picture as a Vermeer than to the biochemist's process of classifying an enzyme as lysozyme. Sir Solly's account is particularly interesting because it catches the subject at the point where it is being converted from an art into a science, and quantification by means of multivariate analysis is replacing the intuitive assessment of experience.

The second half of the book is based on the thesis that decisions about the application of science to social needs must be made politically; here scientists have no more votes than any other group of citizens. Most scientists would, I feel sure, agree with this wholeheartedly and so I imagine would most thinking people. But scientists do have a very definite pedagogical duty. This is to make sure that the political decisions are made in the fullest and clearest knowledge of the scientific background involved and of its limitations. Scientific and technological forecasting is a chancy game because we cannot predict the changes likely in basic science. But even when we choose goals that can be achieved with currently available technology we lack, in a democratic society, the means of obtaining a public consensus if these goals are at all sophisticated. The population explosion is technologically controllable, but is, at least at present, politically unlikely. The health of the community could be improved and the incidence of lung cancer drastically reduced if we abandoned the cigarette, but we are incapable of even fully controlling cigarette advertising. It is not perhaps surprising that Sir Solly thinks that the quality of life may very well decline rather than improve in the years ahead. T. C. WADDINGTON

Critique of Pure Science

The Principles of Scientific Thinking. By R. Harré. Pp. viii + 324. (Macmillan: London, September 1970.) 90s.

HARRÉ points to those obstinate problems in the philosophy of science which surround induction and explanation and argues that they arise from fundamental mistakes—he calls them myths—shared by apparently diverse present day philosophies of science. The myths are most characteristic of logical positivism: those which do not derive from Hume derive from mathematical logic. Among them are Humean atomism: perception gives us knowledge of events occurring at one time and place from which no conclusions about events at other times and places can be deduced; positivism, that to be legitimate a concept must be verifiable in experience; deductivism, which is, in Harré's meaning, the view that there are no necessary connexions except those of deductive logic; and finally, a myth which he might have called verbalism: that scientific thought can be adequately expressed in language, without the aid of pictures and diagrams. Together these principles exclude from science all concern with the hidden constitution of things, yet to explain phenomena is, essentially, to show how they arise from the inner constitution of things, and the acceptability of a theory rests not only upon its conformity with experiment but upon whether it ascribes to things a plausible inner structure. "Those which command serious attention are just the theories which describe models which might be hypostatized to be the actual mechanisms of the nature and the real structure of things." Furthermore, Harré believes that a distinction between causal and accidental regularities can be made by reference to the inner structure of things: causal laws "describe the modes of generation or mechanisms of production of phenomena". In its claim to identify myths lying behind a received doctrine, the introduction to Harré's book bears a striking resemblance to that of Gilbert Ryle's book, *The Concept of Mind*. Perhaps the resemblance is not fortuitous, for the fault Ryle found with Descartes's theory was explanations in terms of occult mechanisms, whereas the fault Harré finds with today's philosophy of science is just the opposite. It forbids the possibility of genuine explanation by restricting science to what is not occult.

The defence of his views leads Harré into discussions of many fundamental problems of the philosophy of science. He is led to the view that nature must have a fundamental structure whose persistence without change demands no

explanation. He attempts an *a priori* rejection of the atomic theory of matter in favour of a field theory and he says that his metaphysical views are closest to those of Faraday.

How successful is this ambitious attempt? There is no doubt that the notion of the inner constitution of things is asked to bear more than it can. It cannot help us to understand causal necessity or to establish, against Hume's arguments, that there is any such thing. Harré's attempt to answer Hume's criticism of the notion is very unconvincing indeed (pages 105–111). Nor, without some change of view about the nature of justification or of knowledge, is the fact that things have an inner constitution much help with the problem of induction. First, it does no more than replace the problem of the justifiability of inductive argument with that of the justifiability of argument by analogy. Second, for basic hypotheses such as some of those of mechanics, no acceptable explanation in terms of inner structure has been proposed to help us choose between the alternative hypotheses left open by induction from experimental instances. Harré is forced to relegate such principles as the law of impact, which stand as firm as any in science, to the status of protolaws. Further, although many scientific explanations do rest upon the inner constitution of things, and Harré is right to feel dissatisfaction with the positivist account of such explanation, it is not clear that all do. The principle of the lever is the basis of many explanations but has no concern with inner mechanisms.

There is one feature of the deductivist philosophy which is regrettably absent from Harré's own. The paradoxes in Hempel's rules of confirmation became apparent because those rules were formulated precisely. Harré not only eschews precise formulation of the principles of analogy which he thinks must replace the principles of induction (page 35), but is prepared to embrace principles, such as affirmation of the consequent, which lead to paradoxes just as serious as those of Goodman and Hempel and much more obvious.

The proof reading has been done very carelessly but, in recompense, there is an extremely useful bibliography.

JOHN WATLING

Tropical Medicine

Tropical Doctor: a Journal of Modern Medical Practice. Volume 1, Number 1. Edited by Dr Hugh Clegg. Quarterly. (Royal Society of Medicine: London, January 1971.) 60s annually; 15s each copy.

THE provision of adequate health services for developing countries is a task of

paramount importance and great difficulty in the face of the continued shortage of professional and auxiliary medical staff. There can be no permanent improvement in the present conditions unless the highest possible priority is given to the training of large numbers of health workers of every description.

Many newly independent nations embarked on the development of full scale medical schools while fully realizing that during the next decade or so, the number of graduates cannot be more than a trickle. But mere numbers of graduates is not enough. There is also the need to maintain the quality of medical practitioners—especially those who work in distant rural areas without an easy access to the medical books and journals that characterizes the present "information explosion". *Tropical Doctor*, fathered, carried to term and skilfully delivered by its editor Dr Hugh Clegg, under the auspices of the Royal Society of Medicine and with the assistance of the Commonwealth Foundation, aims at filling this gap in communication. It is being produced primarily for the benefit of the isolated medical worker who is responsible for a large number of people; he cannot decline this mandate without depriving them of their only source of medical help. Much of the medical practice in the tropics is subject to stagnation because of the remoteness of the under-doctored areas. And yet the man in the field must not only retain his original skill, but must acquire new knowledge of the advancing science and technology applied to medicine and public health.

The first issue of *Tropical Doctor* fulfils brilliantly its promise. The contributors to it come from many parts of the world. Chaudhuri from the School of Tropical Medicine in Calcutta provides an informative paper on the treatment of cholera; Khoo Oon Teik of Singapore deals with lobar pneumonia as seen in the tropics; Burkitt of London outlines the diagnosis and treatment of the tumour that bears his name; Norman from the United Arab Republic writes about hookworm, Wosorun from Accra and Adeyemi from Ibadan discuss the management of burns and head injuries respectively; Cook talks about tropical ulcer and Farman about general anaesthesia.

In the series of special reports there are excellent papers on maternity care in the tropics; on control and eradication of smallpox, on "under fives clinics" and on surgical improvisation in the Pacific. "Newsletters" and replies to "Any Questions" are signed by many distinguished names. There are also two shorter sections on drugs and equipment and finally "News and Notes".

The attractive cover of the journal, its excellent layout, its clear print and

illustrations are bound to enlist many faithful subscribers and readers.

L. J. BRUCE-CHWATT

African Birds

Birds of West Central and Western Africa. By C. W. Mackworth-Praed and C. H. B. Grant. (African Handbook of Birds, Series III, Volume 1.) Pp. xxvii + 671 + 46 plates. (Longman: London, 1970.) 120s.

THE surviving author is to be congratulated on having so nearly completed the ambitious task undertaken 48 years ago; this is the penultimate volume of the six that were planned, and the final one is understood to be well on the way. Captain Grant died in 1958 but had already done much preliminary work even for this last stage. The aim was to provide a work covering all the birds of Africa south of about 20° N latitude. There were to be three series of two volumes apiece, each series constituting an independent regional account. The first dealt with eastern and north-eastern Africa and the second with the southern third of Africa; this one covers west central and western Africa, from southern Mauritania down to the Congo-Zambesi divide in Angola. This area is much more extensive than that covered by either Bannerman's *Birds of Tropical West Africa* (1930–51) or Chapin's *Birds of the Belgian Congo* (1932–54).

The plan inevitably involves substantial overlap between the different series, many species being common to two or all three of the regions. It has thus been necessary, for the sake of consistency, to deal conservatively with changes in nomenclature and new views on taxonomy that have arisen since the earlier volumes were published. The information about individual species and subspecies is on the same lines as before; the stress is on identification and distribution, with brief treatment of chief points of life history and behaviour. There are the same marginal drawings of birds, as well as the colour plates by a number of experienced bird artists; and the same useful distribution maps. Dr W. Serle, with personal experience of this side of the continent, contributes an introductory section on topography and climate.

A. LANDSBOROUGH THOMSON

Antarctic Ecosystems

Antarctic Ecology. Vols. 1 and 2. Edited by M. W. Holdgate. Pp. xx + 604 and x + 607–998. (Academic: London and New York, 1970. Published for the Scientific Committee on Antarctic Research.) 120s and 100s.

THESE are model symposium volumes, well edited and with a strong and coherent theme. They include 81 papers

on all aspects of Antarctic ecology, descriptive, quantitative, behavioural, functional, grouped into fourteen themes, each with a short introduction and a useful edited discussion at the end. The papers are illustrated with photographs, figures and maps, and there are good subject and author indexes at the end of volume two. Volume one deals essentially with marine ecology, volume two with terrestrial and freshwater ecology.

Several of the topics lead into discussions of cropping and conservation of Antarctic ecosystems. Are the krill, supposedly under-exploited by the present decimated whale populations, a potential world food resource, or are they basic to so many Antarctic food chains that it would be hazardous to crop them at all? Cropping at the zooplankton level has always seemed ecologically attractive but mechanically impracticable, but krill are large enough, abundant enough and occur in dense enough shoals to overcome these difficulties. Moiseev suggests that successful exploitation of krill would "probably allow us to double the present catch of aquatic organisms from the world ocean". Other members of the symposium were less sanguine.

What are the hopes for successful conservation of the Antarctic continent, an international resource hitherto almost totally unexploited, with its simplified and presumably vulnerable ecosystems? The Antarctic Treaty, negotiated in 1960, is steadily being endorsed by all the relevant nations and is sometimes hailed as an example of effective international conservation, a model for the internationally agreed world conservation measures essential for man's survival. It is too early to say whether Antarctic conservation is successful and, if so, whether this is because governments have at last seen the light about conservation, or because the economic and/or military advantages of flouting the treaty are negligible. If resources of world fossil fuels become critical and nuclear energy is not an acceptable substitute, will the great Antarctic coalfield be scheduled for exploitation? If so, will the Antarctic Treaty cut much ice?

I would have liked all the maps the same way up. I am surprised the symposium contained only a bare mention of the oft quoted occurrence of organochlorine pesticides in Antarctic animals. A critical discussion of the ecological implications of these results would have been valuable. Few individual scientists will be able to afford *Antarctic Ecology*, though by present standards £11 for one thousand pages is not expensive. All in all, the contributors, publishers and especially the editor are to be warmly congratulated on this *magnum opus* and librarians at least should be encouraged to buy it. It will stand as a major source of reference for many years.

P. J. NEWBOULD

Controlling Parasitism

Recent Advances in Researches on Filariasis and Schistosomiasis in Japan. Edited by Manabu Sasa. Pp. 402. (Tokyo University: Tokyo; University Park: Baltimore, Maryland and Manchester, July 1970.) \$21.

THIS is an account of the work stimulated by the Japan US Cooperative Medical Science Programme which set up a Japanese Panel on Parasitic Diseases in 1965 to study filariasis and schistosomiasis. Medical parasitology as a scientific discipline began with filariasis in 1877, when Patrick Manson discovered that the filarial parasite causing elephantiasis was transmitted by mosquitoes. Filariasis is still a major problem in Asia with more than 200 million people infected. At the time when Manson was working on filariasis in China, Japanese physicians were studying a disease of the liver and bowel that was spreading throughout the Hiroshima prefecture and other parts of southern Japan. The blood fluke causing this disease was eventually discovered by Katsurada in 1904, and he was able to show that it was closely related to the parasite causing bilharziasis in Egypt. Western scientists were completely baffled by this disease and were unable to determine the mode of transmission. The clue came from Japan where Fujinami and others had shown that infection was contracted by bathing in water—Miyairi and Suzuki in 1913 demonstrated that transmission was through a snail host. Professor Yokogawa gives an account of these far reaching discoveries in the section of the book devoted to "Schistosomiasis in Japan"; this is a success story. Japan is one of the few areas of the world where control is effective. In spite of the problem of a large reservoir of infection in animals (which makes control by treatment ineffective), the parasite has been practically eliminated chiefly by killing the snails with molluscicides, and by lining the irrigation canals with concrete to prevent the snails from breeding. In one area the parasite was eliminated when an infected swamp was converted into a golf course.

Elsewhere in the world schistosomiasis is of increasing importance; in Africa and South America practically all new irrigation schemes have been invaded by the snail hosts and control measures are having little effect on the prevalence of the disease. There is no doubt that western scientists and public health workers have still a great deal to learn from their Japanese colleagues when it comes to the control of parasitic diseases. Control is not the only aspect of schistosomiasis dealt with in this book; there are other chapters dealing with the ultrastructure of the parasites, their isoenzymes, the clinical aspects and treat-

ment of the disease and the development of immunological tests.

In the section on filariasis, Professor Sasa and his colleagues show that although this disease has been less effectively controlled than schistosomiasis, there has been a steady decline in the infection rate chiefly as a result of systematic surveys and treatment. As with the work on schistosomiasis there have been numerous supporting studies including observations on spermatogenesis and oogenesis of adult worms using the electron microscope; studies on the dynamics of transmission with a detailed mathematical analysis of epidemiological data; immunological studies with a view to developing better diagnostic techniques; and studies on the periodicity of microfilariae. This latter study takes research on filariasis full circle back to the observations by Manson, who first noted that although the microfilariae are present in the blood in large numbers at night they disappear from the blood during the day. A variety of controlling mechanisms is known but this circadian rhythm is still something of a mystery. Professor Katamine is studying this problem in experimental animals and man in Japan. Of particular interest is the account of the change in rhythm of microfilariae in Japanese emigrants on a two month journey westwards from Japan to Bolivia. This study confirmed that the innate rhythm of the parasites is determined by the sleep rhythm of the host and that there was a gradual shift of peak microfilarial activity in spite of the 13 hour time difference between Japan and Bolivia.

G. S. NELSON

Insect Physiology

An Introduction to Insect Physiology. By E. Bursell. Pp. xiv + 276. (Academic: London and New York, November 1970.) 70s; \$10.

AIMED at the undergraduate and intended as an introduction to physiology, Professor Bursell's book fulfils perhaps the primary requisite of an introductory work in that it can be read easily and completely. As such it contrasts with larger and more comprehensive works such as Chapman's *The Insects, Structure and Function*, and Wigglesworth's *The Principles of Insect Physiology*, which, though excellent works of reference, are at first difficult to grasp as a whole.

Divided into four chief sections, *An Introduction to Insect Physiology* begins with the largest, that concerned with somatic physiology. Here the reader finds the classical subdivisions of physiology, metabolism, digestion, circulation, osmoregulation, excretion and respiration as applied to insects. The second section deals with neuromuscular and

sensory physiology and mechanisms of nervous integration; it also includes a brief but worthwhile digression into the realm of insect behaviour. The third section is devoted to reproduction, development and neuroendocrinology, and section four is entitled, somewhat grandly, "Aspects of Physiological Ecology". Here the temperature and humidity relations of insects have been elevated above their place in more traditional volumes—understandably so because they constitute an important research interest of the author. The possible effects of temperature and humidity on population dynamics are examined in some detail, but the interest of the section lies in the emphasis placed on the difficulties of extrapolation from laboratory conditions to the natural environment, and indeed of the seeming impossibility, with present techniques, of performing experimental biology on field populations of insects.

The task of condensing such a large subject into 276 pages is a daunting one but, as the introduction points out, the method adopted has been to consider the physiological consequences of those features which characterize the insects as a group. Such features are terrestriality, small size, capability for flight and possession of an exoskeleton. Well aware of the difficulties of such a task, the author is at times over-apologetic; the frequency of introductory and concluding remarks is at times tiresome, although their value in reinforcing points made is undoubted.

The text consists of generalizations, illustrated with well chosen and, for the most part, very up to date examples and brevity has not destroyed the sense of critical approach and scientific currency which help to sustain interest. As one might expect from the price, this is a glossy book, much in the vein of other Academic Press publications, and the excellent text figures are almost entirely drawn from source literature.

Here then is a genuinely introductory account which is informative, concise, and about as readable as any account of the subject can be.

JOHN A. PATTERON

The Opossum Brain

The Brain of the Opossum (Didelphis marsupialis): A Cytoarchitectonic Atlas in Stereotaxic Coordinates. By E. Oswald-Cruz and C. E. Rocha-Miranda. Pp. 99 + 45 plates. (Instituto de Biofísica, Universidade Federal do Rio de Janeiro: Rio de Janeiro; Livingstone: Edinburgh, 1968.) 280s.

This work is a useful addition to others now available providing illustrations of a standard series of brain sections together with stereotaxic coordinates

based on the Horsley-Clarke orientation system. Their chief value is to neuro-anatomists and physiologists for the accurate placing of small lesions or the positioning of electrodes with minimal operative interference. They have a secondary value in providing a standard atlas and terminology for the particular brain dealt with which can be helpful in any neurological investigation requiring the identification of nuclei or of anatomically defined groups of nerve cells.

The present work, like most of the others, is limited to the illustration of cytoarchitectural features, although the positions of a number of important fibre systems are indicated and fibre-stained preparations were studied by the authors. It deals with two sub-species of *Didelphis marsupialis*, *aurita* and *virginiana*, the opossums of South and North America respectively. Anatomically the brains are so alike that both are adequately illustrated by the series of 45 transverse sections which are reproduced from the one sub-species, *D. marsupialis aurita*. A simple method for the correction of the given coordinates, necessitated by differences in animal sizes, is fully described. In preparing the illustrations of the standard brain, every care appears to have been taken to apply further corrections for errors introduced by shrinkage during fixation, compression during sectioning and small variations in plane of section.

Some 200 cytoarchitectural areas, nuclei and the like, are labelled, and a synoptic description is given of each. These descriptions are limited to relatively simple features such as cell size and form, and comparatively few references are given to more detailed descriptions in the literature. A bibliography which is comprehensive for the species under study is provided. Although the chief areas of the cerebral cortex are indicated in some of the sections, they are not described. Principal attention is given to subcortical features, as is natural in a work intended primarily for guidance when stereotaxic methods are being employed and the direct visual placement of lesions or electrodes in the brain is impracticable.

Obviously, this work will be of great value in North and South America where the opossum is readily available as an experimental animal. Its secondary value as a well illustrated atlas of an important primitive mammalian brain, to which much neuroanatomical attention has been directed, is not negligible. From a descriptive point of view, however, it adds little to what can be learnt from accounts in well known and easily accessible neurological journals. Although a useful illustrated introduction to such literature, it is doubtful if the cost could be justified for this purpose alone.

F. GOLDBY

Elastic Waves

Crystal Acoustics: Introduction to the Study of Elastic Waves and Vibrations in Crystals. By M. J. P. Musgrave. Pp. xv + 288. (Holden-Day: San Francisco, 1970.) 230s.

EXPERTS on the theory of elastic waves are not greatly to the fore as writers of textbooks. Apart from Kolsky's monograph *Stress Waves in Solids* (first published in 1953 and concerned only in part with elastic waves), there is little in the way of systematic expositions suitable, say, for final year undergraduate and postgraduate students. Yet the interest of the subject is widespread, solid state physics, geophysics, various branches of engineering, and theoretical mechanics being only some of the disciplines drawing upon existing knowledge of elastic wave theory and, in varying degrees, adding to it.

A text of modest scope has little chance of satisfying all these varied needs and Dr Musgrave has wisely planned his book within clearly defined limits. The central topic is the theory of elasticity as applied to wave motions and vibrations of anisotropic materials with symmetry properties representative of crystal structure. Effects associated with non-linearity, thermo-mechanical and acoustic-electromagnetic interactions and non-elastic behaviour receive only passing mention, and the theory of elastic wave propagation in isotropic materials is treated as a degenerate case meriting no special emphasis. Experimental results are freely reported but there is no discussion of experimental methods as such.

Part one consists of sixteen shortish chapters developing the theory of elasticity for small deformations, collecting together relevant facts from crystal physics, and then providing a well-organized account of basic properties of small amplitude waves propagating in anisotropic materials with particular reference to media possessing symmetries appropriate to the most commonly encountered crystal classes. Embedded in these chapters is a connected account of the important contributions to elastic wave theory which the author has himself published.

Part two, made up of four chapters on the dynamics of lattice vibrations and its application to the construction of models for cubic crystals, reflects Dr Musgrave's interest in the physics of crystalline materials. This is an attractively written introduction to a subject which will be unfamiliar to readers whose approach to crystal deformation has been exclusively from the viewpoint of continuum mechanics. The book as a whole fills a gap in the textbook literature; one regrets that the financial deterrent to would-be purchasers is so severe.

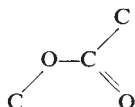
P. CHADWICK

CORRESPONDENCE

Molecular Models

SIR,—Atomic model building (or, as some might say, guessing) is a powerful tool in determining the structures of molecules. In the cases of the α -helix and DNA, for example, the structures would not have been proposed without the aid of accurate model building. In both cases, however, the model building was backed up by sound stereochemical knowledge. We would like to draw your attention to the most important stereochemical error that occurs in the recent article in which W. H. Beers and E. Reich use model building to relate the structure of acetylcholine to its activity as a neurotransmitter¹.

All five atoms of an ester group



are coplanar due to the partial double bond character of the O—C bond (ref. 2). The group has a resonance energy of about 24 kcal per mole (ref. 2) and this energy must be added for any large rotation to take place about the O—C bond.

Beers and Reich do not describe their model for acetylcholine in the text of the article, but inspection of Figs. 2 to 6 shows the molecule, $(\text{CH}_3)_3\text{N}^+\text{CH}_2\text{CH}_2\text{OCOCH}_3$, with a rotation about the O—C bond of approximately 90° from the coplanar conformation. Such a conformation, at 24 kcal per mole above the ground state, would occur for less than one molecule in 10^{15} . As less than 10^8 molecules are released for the transmission of one nerve impulse³ it seems doubtful that this model of acetylcholine is relevant to the molecule's role as a neurotransmitter.

Yours faithfully,

CYRUS CHOTHIA

MRC Laboratory of Molecular Biology,
Hills Road,
Cambridge

PETER PAULING

Department of Chemistry,
University College London,
Gower Street,
London WC1

¹ Beers, W. H., and Reich, E., *Nature*, **228**, 917 (1970).

² Pauling, L., *The Nature of the Chemical Bond*, 3rd ed. (Cornell University Press, Ithaca, 1960).

³ Katz, B., *Nerve, Synapse and Muscle*, 120 (McGraw-Hill, London, 1966).

Electrical Plant

SIR,—May I comment on the statements concerning the unreliability and breakdown of 500 MW and 660 MW generating units and the way the Central Electricity Generating Board manages its affairs (*Nature*, **228**, 1126 and 1246; 1970).

The Select Committee on Science and Technology (HMSO, 1970) concluded that there were welding defects and hair-line cracks in the boilers. The CEGB states¹ that "the problem in this country is accentuated by the high proportion of new plant on the system".

Could it be that too much attention has been given to designing plant which will operate at an excessively high steam temperature without an adequate regard for plant reliability?

In the United States there are 25 units² under construction of between 500 MW and 1,380 MW with a maximum steam temperature of $1,010^\circ\text{F}$ which is some 40°F below the CEGB units which are under construction.

Yours faithfully,

F. H. DENNIS

12 Cintra Park,
London SE19

¹ CEGB Annual Report and Accounts, 1969–70, 3 (1970).

² *Electrical World*, **174**, No. 8, 35 (1970).

Acanthaster in the Indian Ocean

SIR,—Having dived off the east coast of Africa since 1959, we very rarely saw *Acanthaster planci* (crown of thorns starfish) until 1968. One specimen was seen in 1961 at Bazaruto Island.

In January 1968 we spent some time in the Bay of Nacala ($14^\circ 25'\text{S}$, $40^\circ 40'\text{E}$), northern Mozambique. This is being developed as a harbour but we saw no evidence of blasting or dredging¹. Large areas of the reef were dead or apparently dying. In retrospect we have thought that this was probably attributable to *A. planci*² of which we saw and photographed a great number. At the time we were unaware of the threat that this starfish was presenting in the Pacific Ocean^{1–3}.

The following year, while visiting several islands in the Indian Ocean, we saw vast areas of devastated coral and many specimens of *A. planci* at Juan de Novo (17°S , 42.5°E). This is a small, totally isolated island with a Melanesian

population of about fifteen and with fewer than this number of visitors in the last two years. A passageway through the reef had been blasted on the leeward side of the island to allow small boats through the coral to load phosphates mined on the island. This activity had stopped about three years previously.

We are happy to report that we saw no evidence of *A. planci* or coral damage at either Mayotte in the Comores or at Aldabra.

In December 1969 we saw about ten specimens in six half-hour^{1,4} dives at Bazaruto Island (22°S , 36°E). Some of these were seen at Punto Dundo, a rocky outcrop with very little coral, between Bazaruto and Benghuerra. At this point the tidal current reaches 5–7 knots. Others we saw at Gengareme. This is a sheltered bay on the lee of the island where there are mainly large brain corals (lobophyllia) isolated by sand. Shortly after this, another expedition reported collecting sixty³ specimens in one dive from the reef two miles NNE of Punto Dundo. This reef has abundant and varied coral growth, and is subjected to perpetual breaking water. There was no evidence of coral damage in any of these three areas.

Another threat to the life on coral reefs is becoming more and more evident in Northern Mozambique. The local fishermen have learnt to use goggles and spearguns and indiscriminately shoot all coral fish over about three inches long. This results in a dearth of fish above a depth of about thirty-five feet. The fish deeper than this are very shy. We feel that concerted effort is required to protect life on coral reefs, such as at Watamu, Kenya and Aldabra.

Yours faithfully,

T. P. BLIGH
N. BLIGH

PO Box 809,
Johannesburg,
South Africa

¹ Cheser, R. H., *Science*, **165**, 280 (1969).

² *Australian Academy of Science, Report No. 11* (1970).

³ Dixon, B., *New Scientist*, Oct. 30, 226 (1969).

⁴ Vine, P. J., *Nature*, **228**, 341 (1970).

Nonsense Fragments

SIR,—The December 19 issue of *Nature* contains two articles and an editorial review^{1–3} concerned with the fate of fragments of proteins produced by *E. coli* nonsense mutants. The authors claim

to have demonstrated the existence of a mechanism, presumably enzymatic, causing the rapid degradation of fragments *in vivo*, and discuss at some length the physiological implications of this finding. Neither the authors nor the reviewer consider an alternative explanation, that the fragments might have been released from the bacteria into the medium and in this way have escaped detection by the methods used. There is in fact some evidence for this explanation, from earlier experiments with alkaline phosphatase nonsense mutants of *E. coli*; phosphatase fragments were found to be rapidly released into the medium, an average time of about 20 minutes being required for a fragment to appear in the medium after being synthesized intracellularly^{4,5}. The behaviour of phosphatase fragments could represent a special case, because the phosphatase enzyme in a standard strain of *E. coli* is localized in the region between the outer cell wall and the inner membrane (the periplasmic space); consequently phosphatase fragments might escape from a cell more readily than fragments of other proteins. Nevertheless, in the absence of evidence to the contrary, the intracellular disappearance of fragments of any protein might result, not from degradation, but rather from the passage of the fragments into the surrounding medium. Until this possibility has been tested, the conclusion that *E. coli* protein fragments are degraded *in vivo* must be regarded as speculative.

Yours faithfully,

ALAN GAREN

Department of Molecular Biophysics
and Biochemistry,
Yale University,
New Haven, Connecticut

¹ Goldschmidt, R., *Nature*, **228**, 1151 (1970).

² Platt, T., Miller, J. H., and Weber, K., *Nature*, **228**, 1154 (1970).

³ *Nature*, **228**, 1137 (1970).

⁴ Suzuki, T., and Garen, A., *J. Mol. Biol.*, **45**, 549 (1969).

⁵ Natori, S., and Garen, A., *J. Mol. Biol.*, **49**, 577 (1970). Reference to the loss of phosphatase fragments from cells is specifically made on page 587 of the second article, and the experimental details have been submitted for publication in *J. Mol. Biol.*

This letter has been shown to our correspondent, who replies:

The idea that cells might extrude unwanted nonsense fragments is appealing, but seems unlikely to apply for β -galactosidase and lactose repressor mutants. Neither of these proteins is located in the periplasmic space; this must imply that a special (enzyme) system would be needed to transport the nonsense fragments out of the cell. This seems more complicated than merely degrading the fragments, and would in any case demand the type of recognition mechanism which the authors discussed to discriminate mutant from normal proteins. An extrusion mechanism does not, therefore, seem to possess any theoretical advantage, and experimentally Goldschmidt was able to

detect and identify a protein fragment which is probably produced by the postulated endopeptidase degradation of the β -galactosidase nonsense fragment.

Neuromythology?

SIR,—Is the large daily loss of neurones from the brain, which figures in all the textbooks and provides the basis for Dawkins's¹ ingenious model of memory, as well established as he suggests? I write because the matter, which was the subject of a fair amount of controversy at a recent American research course on ageing, is of considerable importance for models of psychological and psychiatric ageing as well.

Leaving aside insect work, the prime authority for a massive loss is Brodie², whom Dawkins quotes, though the original mammalian claim was made by Hatai³ for the rat, and repeated by Vogt and Vogt⁴. The loss of Purkinje cells from rat brain found by Inukai⁵ has not been discovered in ageing hamsters⁶. With regard to the cortex, which would appear to be the critical site for Dawkins's purpose, some loss is reported⁷ but in the rat it appears to be trifling⁸. The most recent title indicates no loss whatever with age from the human cochlear nucleus⁹.

In view of the importance of the subject and the categorical nature of the textbook statement, the matter of cell loss with age among fixed postmitotics is due for careful re-examination, in spite of the tiresome task of cell counting which it involves. It would be of interest to know whether studies of this kind in mammals are already in progress.

Yours faithfully,

ALEX COMFORT

Director of Research, Gerontology,
University College London

¹ Dawkins, R., *Nature*, **229**, 118 (1971).

² Brodie, H., *J. Comp. Neurol.*, **102**, 511 (1955).

³ Hatai, S., *J. Comp. Neurol.*, **12**, 107 (1902).

⁴ Vogt, C., and Vogt, O., *Nature*, **158**, 304 (1946).

⁵ Inukai, T., *J. Compar. Neurol.*, **45**, 1 (1928).

⁶ Wilcox, H. H., *J. Gerontol.*, **11**, 442 (1956).

⁷ Wright, E. A., and Spink, J. M., *Gerontologia*, **3**, 277 (1959).

⁸ Brizzee, K. R., Sherwood, N., and Timiras, P. S., *J. Gerontol.*, **23**, 289 (1968).

⁹ Konigsmark, B. W., and Murphy, E. A., *Nature*, **228**, 1355 (1970).

ESRO Satellite

SIR,—It was not our intention to imply that a satellite needs to be stabilized to the same accuracy with which one desires to locate the X-ray sources. There is, however, a relation between the two as illustrated by the example of Sco X-1 given by Dr Gursky¹. As he correctly states, the requirement is to know, with high precision, the attitude, at the same time maintaining a stability commensurate with the field of view of the

instrument. For the HEAO mission to which Dr Gursky refers (that carrying modulation collimators, not the large X-ray telescope planned for a later mission) he claims a limiting accuracy in location of several arc seconds.

If this can be achieved, the two missions would give complementary results in the location of sources since the occultation satellite, although in principle capable of higher precision, locates a smaller fraction of the observed sources. On the other hand, an important characteristic of the occultation method is that the angular dimensions and relative structure of extended sources can be obtained with even higher accuracy than that of location, down to small fractions of an arc second.

We should finally note that the terms "complex" and "expensive" are, of course, relative rather than absolute, and should be read in the context of the ESRO budget which does not at present envisage satellites of the HEAO type.

Yours faithfully,

J. ORTNER

The ESRO Mission Definition Group
for the Highly Eccentric X-ray
Astronomy Lunar Occultation Satellite

¹ Gursky S., H., *Nature*, **228**, 1121 (1970).

Neural Nomenclature

SIR,—In 1933, Dale¹ suggested the terms "adrenergic" and "cholinergic" for the two types of autonomic nerve fibre known at that time. He introduced these terms "to assist clear thinking, without committing us to precise chemical identifications, which may be long in coming".

In the early 1960s powerful nerves were found to supply that gut which were neither cholinergic nor adrenergic²⁻⁴. Since then, there have been a number of reports concerning details of their distribution, structure and function⁵⁻⁷, but their description as non-adrenergic, non-cholinergic nerves is clumsy and somewhat negative. Evidence has recently been presented that the transmitter substance released from these nerves may be ATP or some related purine nucleotide⁸. It would therefore seem suitable, for the same reasons put forward by Dale in 1933, to propose that the new nerves be termed "purinergic".

Yours faithfully,

G. BURNSTOCK

Department of Zoology,
University of Melbourne, Parkville, 3052,
Victoria, Australia

¹ Dale, H. H., *J. Physiol.*, **80**, 10 (1933).

² Burnstock, G., Campbell, G., Bennett, M., and Holman, M. E., *Nature*, **200**, 581 (1963).

³ Burnstock, G., Campbell, G., Bennett, M., and Holman, M. E., *Intern. J. Neuropharmacol.*, **3**, 163 (1964).

⁴ Burnstock, G., Campbell, G., and Rand, M. J., *J. Physiol.*, **182**, 504 (1966).

- ⁵ Burnstock, G., *Pharmacol. Rev.*, **21**, 247 (1969).
⁶ Campbell, G., in *Smooth Muscle* (edit. by Bülbring, E.), 451 (Edward Arnold, London, 1970).
⁷ Burnstock, G., and Iwayama, T., in *Progress in Brain Research* (Elsevier, Amsterdam, in the press).
⁸ Burnstock, G., Campbell, G., Satchell, D. G., and Smythe, A., *Brit. J. Pharmacol.*, **40**, 11 (1970).

Milliard or Gillion?

SIR,—When a word has become ambiguous it should be discarded. Attempts

to reimpose precision could ultimately be successful, but there will be an inconvenient phase in which, though a writer may know what the word should mean, there is no certainty that readers will be in the same happy state. The word billion seems to be irrevocably lost. Boroughs's suggestion (*Nature*, **229**, 142; 1971) that the vague word "milliard" should be defined as 10^9 is unsatisfactory because it is already current and readers will not, for a time, know whether it is being used precisely or not. May I repeat the suggestion

(*Nature*, **220**, 312; 1968) that we should make use of the lucky accident that M is both the internationally agreed symbol for 10^6 and the initial letter of million. G and T are the symbols for 10^9 and 10^{12} and could be used to form the new and therefore unambiguous words gillion and tillion.

Yours faithfully,

N. W. PIRIE

42 Leyton Road,
Harpenden,
Hertfordshire

Obituary

Professor A. S. Besicovitch

ABRAM SAMOILOVITCH BESICOVITCH, Rouse Ball professor of mathematics at Cambridge from 1950 to 1958 and a Fellow of Trinity College, died on November 3, 1970.

He was born in 1891 and studied at St Petersburg, later teaching there and at Perm. He left the USSR in 1925 and worked in Copenhagen with Harald Bohr, who had recently built up his theory of continuous almost-periodic functions. In collaboration with Bohr and independently, Besicovitch enlarged the concept of almost periodicity to wider classes of functions. With an appropriate definition, a generalized almost-periodic function possesses a Fourier series and it satisfies Parseval and Riesz-Fischer theorems and also a Cantor theorem of uniqueness. Besicovitch brought all these investigations into a book in 1932.

Besicovitch was one of the most powerful mathematical analysts of his generation. He has been described as a master of intricate construction. An early illustration of this gift was his example—published in 1922 in a Russian journal which did not reach other countries—of

a plane set made up of linear segments of length 1, one in every direction, having zero Jordan measure. He saw later that this set could be adapted to solve Kakeya's problem of finding the figure of least area in which a segment of unit length can be rotated through a complete revolution. By rotating a pencil on a table, the reader may guess that the figure sought is a three-cusped hypocycloid, having area $\pi/8$. The paradoxical truth is the existence of figures of arbitrarily small area.

In the 1930s Besicovitch wrote many papers on plane sets of points which have finite length in the sense of Carathéodory's linear measure. The density of such a set E at a point x can be defined by the limit of the linear measure of the part of E within a circle centre x divided by the diameter of the circle. Sets may be regular or irregular. A regular set behaves like a rectifiable curve, having a tangent at almost every point. An irregular set has positive density in every sector drawn from almost every one of its points. The first proofs of such theorems were long and complicated and they exemplify one of Besicovitch's aphorisms, "A mathematician's reputation rests on his bad proofs". In later

papers he replaced some of his pioneering bad proofs by more elegant good proofs.

Hausdorff measure was a topic to which Besicovitch returned repeatedly, using the concept to refine results about sets of points and real functions. Suppose, for instance, that x is a number for which infinitely many rationals m/n exist differing from x by less than $1/n^q$, where q is less than one; then the set of x has Hausdorff dimension $2/q$.

In 1945 Besicovitch's gift for intricate construction led him, to his own surprise, to another paradox. He defined in space a surface homeomorphic with a disk (or a sphere), which has three-dimensional measure equal to 1 (say) but arbitrarily small surface area in the Lebesgue-Fréchet sense of the lower bound of approximating polyhedra. This led him to the view that results based on the Lebesgue-Fréchet notion of area must be recast in terms of Hausdorff plane measure. He and his pupils carried out a large part of this programme.

In trying to convey an impression of Besicovitch's magnificent mathematical gifts, no one could forget that they were matched by gifts of humanity which endeared him to pupils and friends and especially to young children.

Announcements

University News

Professor G. Slaney has been appointed to the Barling chair of surgery, University of Birmingham, in succession to Professor A. L. d'Abreu who retires this year. Professor O. L. Wade has been appointed to the newly established chair of clinical pharmacology and Dr E. H. Ashton to a chair of anatomy established for one tenure only. The title of professor of reproductive endocrinology has been conferred on Dr P. Eckstein.

Dr S. R. Stitch has been awarded a personal professorship in steroid endocrinology by the University of Leeds.

Dr H. Smith, University of Manchester, has been appointed to the chair of plant physiology in the Faculty of Agricultural Science, University of Nottingham.

Dr A. T. Cowie has been appointed head of the Physiology Department, National Institute for Research in Dairying, University of Reading, in succession to the late Professor Folley.

Appointments

Professor Alwyn Williams, Queen's University of Belfast, has been appointed a trustee of the British Museum (Natural History), in succession to Professor O. M. B. Bulman.

Dr Philippe Shubik, Eppley Institute for Research into Cancer, University of Nebraska, and James S. Gilmore, jun.,

Gilmore Broadcasting Corporation, Kalamazoo, have been appointed to the National Advisory Cancer Council of the US National Institutes of Health.

The following have been appointed vice-presidents of the Royal Society: Sir Frederick Bawden, Rothamsted Experimental Station; Sir Bernard Katz, University College London; Sir Harrie Massey, University College London; Sir Harold Thompson, University of Oxford; Professor W. R. S. Doll, University of Oxford; Professor F. Hoyle, University of Cambridge; Dr J. M. Menter, Tube Investments Limited.

Miscellaneous

Professor P. M. Maitlis, McMaster University, has been awarded the 1970

Steacie prize in the natural sciences for his work in organometallic chemistry.

Professor Frank Press, Massachusetts Institute of Technology, and **Sir Richard Woolley**, astronomer royal, have been awarded gold medals of the **Royal Astronomical Society**. The Eddington medal has been awarded to **D. G. King-Hele** and the Jackson-Gwilt medal to **A. W. J. Cousins**.

ERRATUM. In the article "Effect of Mineral Adjuvant on Lymphocyte Cooperation in the Secondary Antibody Response to a Hapten-Protein Conjugate" by Samuel Strober (*Nature*, **228**, 1324; 1970), the word "deoxynucleoprotein" in line 4 should read "dinitrophenol". The description of the article on the contents page should have stated that mineral adjuvant obviates, not enhances, lymphocyte cooperation in the secondary antibody response to a hapten-protein conjugate.

ERRATUM. The price of the book *The California Earthquake of April 18, 1906* (Carnegie Institution of Washington) which was reviewed in the December 19 issue of *Nature* (**228**, 1231; 1970) is \$12.50 and not \$87 as was stated.

ERRATUM. In the review entitled "Views of Deafness" by J. D. Hood (*Nature*, **229**, 68; 1971), the first sentence should have read "Sensorineural hearing loss is the name given to deafness resulting either from some disorder of the inner ear, or, more rarely, from a lesion of the central nervous pathways to the brain".

International Meetings

March 9–10, **Appendages of the Skin**, Eastbourne (Society of Cosmetic Chemists of Great Britain, 56 Kingsway, London WC2).

March 15, **Collagen Club Meeting**, Cambridge (Dr C. I. Levene, Dunn Nutritional Laboratory, Milton Road, Cambridge).

March 16–18, **Electrical Safety in Hazardous Environments**, London (Institution of Electrical Engineers, Savoy Place, London WC2R 0BL).

March 22–24, **Biomathematics and Computer Science in the Life Sciences**, Houston (Office of the Dean, University of Texas Graduate School of Biomedical Sciences at Houston, Division of Continuing Education, PO Box 20367, Houston, Texas 77025, USA).

March 23, **Radiation Hazards**, London (Professor J. H. Martin, Department of Medical Biophysics, The University, Dundee DD1 4HN).

March 30–April 1, **Electrochemical Engineering**, Newcastle upon Tyne

(ECES, 16 Belgrave Square, London SW1).

March 30–April 2, **Fluid Sealing**, Coventry (H. S. Stephens, 5th ICFS, British Hydromechanics Research Association, Cranfield, Bedford).

April 15–16, **Industrial Innovation through Contract Research**, Newcastle upon Tyne (T. W. Ditchburn, University of Newcastle upon Tyne, 6 Kensington Terrace, Newcastle upon Tyne NE1 7RU).

April 18–23, **Solvent Extraction**, The Hague (ISEC 71, 14 Belgrave Square, London SW1).

April 22–23, **Annual Congress and Scientific Exhibition of the British Institute of Radiology**, London (British Institute of Radiology, 32 Welbeck Street, London W1M 7PG).

April 26–28, **Motorways in Britain Today and Tomorrow**, London (Conference Department, Institution of Civil Engineers, Great George Street, London SW1).

April 28–30, **Modern Steam Plant Practice**, The Hague (Institution of Mechanical Engineers, 1 Birdcage Walk, Westminster, London SW1).

April 28–May 2, **Protides of Biological Fluids**, Bruges (Dr H. Peeters, Simon Stevin Instituut, Jerusalemstraat 34, B.8000, Brugge, Belgium).

May 16–21, **Biological Significance of Transplantation Antigens**, Rovinj, Yugoslavia (Dr J. G. Howard, Department of Experimental Immunobiology, Wellcome Research Laboratories, Langley Court, Beckenham, Kent).

May 18–21, **Debating Electronics**, London (Industrial Exhibitions Ltd, 9 Argyle Street, London W1V 2HA).

June 14–18, **European Association of Radiology Congress**, Amsterdam (Secretariat, c/o Holland Organization Centre, 16 Lange Voorhout, The Hague, Holland).

June 15–17, **Textiles for Comfort**, Manchester (Mr J. K. Jackson, Shirley Institute, Didsbury, Manchester M20 8RX).

June 21–25, **Oil and Colour Chemists' Association Technical Exhibition**, London (R. H. Hamblin, Oil and Colour Chemists' Association, Wax Chandlers' Hall, Gresham Street, London EC2V 7AB).

June 29–July 16, **Geophysical Fluid Dynamics** (summer school), Bangor (Dr S. A. Thorpe, National Institute of Oceanography, Wormley, Godalming, Surrey).

July 5–24, **Our Buildings (Shells) and the Human Settlements**, Athens (P. Psomopoulos, International Programs, Athens Center of Ekistics, Box 471, Athens, Greece).

July 12–16, **Time of Flight Mass Spectrometers**, Salford (Dr D. Price, Department of Chemistry and Applied Chemistry,

University of Salford, Salford, Lancashire M5 4WT).

July 12–16, **Enzymes and their Use in Analysis and Clinical Diagnosis**, Cambridge, Massachusetts (Director of the Summer Session, Room e19-356, Massachusetts Institute of Technology, Cambridge, Massachusetts 02139, USA).

July 19–22, **Endocrine Polypeptide Hormones and their Secreting Cells**, London (Dr G. V. Foster, Endocrinology 1971, Royal Postgraduate Medical School, Ducane Road, London W12).

August 16–September 3, **Differential Geometry, Differential Topology and Applications**, Halifax, Nova Scotia (Canadian Mathematical Congress, 985 Sherbrooke Street West, Montreal 110, Quebec, Canada).

August 23–27, **Medical and Biological Engineering**, Melbourne (Dr David Dewhurst, Department of Physiology, University of Melbourne, Parkville, Victoria, Australia 3052).

September 5–12, **Conference of Women Engineers and Scientists**, Turin (Dr A. E. Amour, Corso Vinzaglio 14, 10121 Torino, Italy).

September 6–8, **Photochemistry**, Bordeaux (Professor J. Jousset-Dubien, Faculté des Sciences de Bordeaux, 351 Cours de la Liberation, 33 Talence, France).

September 7–10, **Organic Geochemistry**, Hannover (Dr H. Wehner, Bundesanstalt für Bodenforschung, 3 Hannover-Buchholz, Postfach 54, Germany).

September 8–9, **High Voltage Insulation in Vacuum**, London (Meetings Officer, The Institute of Physics and the Physical Society, 47 Belgrave Square, London SW1).

September 9–11, **Photosensitization in Solids**, Sarlat, Dordogne (Jean Bourdon, Kodak-Pathe, Centre de Recherches, 94 Vincennes, France).

September 20–25, **Astronautical Congress**, Brussels (Mr P. Contensou, International Astronautical Federation, 250 Rue Saint-Jacques, 75 Paris 5, France).

September 27–October 1, **Automatic Control and Computers in the Medical Field**, Brussels (Secretariat de l'IBRA, 3 rue Ravenstein, B.1000 Brussels, Belgium).

October 4–6, **Turbulence in Liquids**, Rolla (G. K. Patterson, Department of Chemical Engineering, University of Missouri-Rolla, Rolla, Missouri 65401, USA).

October 24–29, **Aviation and Space Medicine**, Tel Aviv (Dr I. Glazer, El Al Israel Airlines, Lod Airport, Israel).

November 2–4, **Salinity and Water Use**, Canberra (Dr T. Talsma, CSIRO Division of Plant Industry, PO Box 109, Canberra City, ACT 2601, Australia).

British Diary

Monday, January 25

Changes and the Future in Electrical Engineering (7.30 p.m.) Mr S. Z. de Ferranti, Institution of Electrical Engineers, at the Free Trade Hall, Manchester. (Faraday Lecture.)

Computer Graphics and the Electrical Engineer (6.30 p.m.) Mr M. Clayton and Mr D. W. Davis, Institution of Electrical Engineers, at Liverpool Polytechnic, Byrom Street, Liverpool.

Concorde Electronics (6.30 p.m.) Mr H. Hill, Institution of Electrical Engineers, at Salisbury and South Wiltshire College of Further Education, Salisbury.

The Mechanism of Genetic Recombination (5.30 p.m.) Dr H. L. K. Whitehouse, University of London, in the Anatomy Lecture Theatre, University College London, Gower Street, London WC1.

Tuesday, January 26

Consideration of the Relative Values of True and Infrared Colour Aerial Photography for Geological Purposes, Dr J. A. E. Allum; **Geology and Evaluation of Placer Gold Deposits in the Klondike Area, Yukon Territory**, Mr B. W. Hester; **An Inter-Laboratory Survey of the Accuracy of Ore Analysis**, Mr B. Lister and Dr M. J. Gallagher (5 p.m.) Institution of Mining and Metallurgy, at the Geological Society, Burlington House, Piccadilly, London W1.

Control Equipment Systems Design in the United Kingdom Power Industry (conference) Institution of Mechanical Engineers, at 1 Birdcage Walk, London SW1.

Decimalization—the Eighteenth-Century Origins (1.20 p.m.) Dr W. A. Smeaton, University of London, in the Botany Theatre, University College London, Gower Street, London WC1.

Electronics—Its Future in Navigation (7.30 p.m.) Mr F. S. Stringer, Institution of Electrical Engineers, at Livingstone Tower, Strathclyde University, Glasgow. (Silvanus P. Thompson Lecture.)

L-dopa Metabolism in Man (5.30 p.m.) Dr M. Sandler, University of London, at the Institute of Child Health, 30 Guilford Street, London WC1. (Sixth of fifteen lectures on "The Scientific Basis of Medicine" organized by the British Postgraduate Medical Federation.)

Metallurgical Problems at the CEEB (7.45 p.m.) Mr M. G. Gemmil, Leeds Metallurgical Society, at the Houldsworth School of Applied Science, The University, Leeds.

The Application of Fracture Mechanics to Fatigue Crack Propagation (6 p.m. discussion) Institution of Mechanical Engineers, at 1 Birdcage Walk, London SW1.

The Chemical Control of Plant Growth (5.30 p.m.) Professor R. L. Wain, FRS Royal Institution, at 21 Albemarle Street, London W1. (Lecture for Sixth Form Pupils from Schools in London and the Home Counties. To be repeated on January 27, February 2 and 3.)

The Design and Construction of the Ocean Terminal, Hong Kong (5.30 p.m.) Mr S. E. Faber, Mr J. C. Faber and Mr J. M. Thomas, Institution of Civil Engineers, at Great George Street, London SW1.

The Open University (7 p.m.) Professor J. J. Sparkes, Institution of Electrical Engineers, at the North Staffordshire Polytechnic, Stafford.

The Role of Industry in the Training of Undergraduate Engineering Students (10.30 a.m. colloquium) Institution of Electrical Engineers, at Savoy Place, London WC2.

Wednesday, January 27

Discussion Meeting (6.30 p.m.) Society for Analytical Chemistry, Microchemical Methods Group, at Imperial College, London SW7.

Electronic Techniques in Archaeology (6.45 p.m.) Mr M. J. Aitken, Institution of Electrical Engineers, at Renold Building, UMIST, Manchester. (Silvanus P. Thompson Lecture.)

Management of RAF Electronic Engineering Projects (6 p.m.) Institution of Electronic and Radio Engineers, at 9 Bedford Square, London WC1.

Robert Boyle's "Essays of Effluviuims" 1673 (1 p.m.) Mr L. R. Day, Royal Institution, History of Science Discussion Group, at 21 Albemarle Street, London W1.

The Evaporation of Heat Sensitive Food-stuff Liquids (10 a.m. symposium) Society of Chemical Industry, Food Group—Food Engineering Panel, in the Lecture Theatre of the Zoological Society of London, Regent's Park, London NW1.

The Use and Abuse of Materials in Ocean Engineering (6 p.m.) Dr D. Birchon, Institution of Mechanical Engineers, at 1 Birdcage Walk, London SW1. (Thomas Lowe Gray Memorial Lecture.)

Thursday, January 28

Flight Recording (6.30 p.m.) Mr R. Parsons, Institution of Electronic and Radio Engineers, jointly with IEE, at the University Engineering Laboratories, Trumpington Street, Cambridge.

Angiotensin in the Central Control of Automatic Function (5.30 p.m.) Dr R. D. Lowe, University of London, at the Institute of Child Health, 30 Guilford Street, London WC1. (Seventh of fifteen lectures on "The Scientific Basis of Medicine" organized by the British Postgraduate Medical Federation.)

Laser Applications in Electronics (7 p.m.) Professor W. A. Gambling, Institution of Electrical Engineers, jointly with IERE, at the Abbey Hotel, Malvern.

Magnetism, Parts 1, 2 and 3 (1 p.m. film) Royal Institution, at 21 Albemarle Street, London W1.

Tower Clocks in Denmark (6 p.m.) Dr Hans Stiesdal, Royal Institution, jointly with the Antiquarian Horological Society, at 21 Albemarle Street, London W1.

Friday, January 29

Kinetic Determinations of Bond Dissociation Energies (1 p.m.) Dr R. Walsh, Royal Institution, Photochemistry Discussion Group, at 21 Albemarle Street, London W1.

The Proper Study of Mankind (9 p.m.) Sir Mortimer Wheeler, FRS, Royal Institution, at 21 Albemarle Street, London W1.

Saturday, January 30

Whales and Whaling (3.30 p.m.) Dr Robert Clarke, Inner London Education Authority, at the Horniman Museum, London Road, Forest Hill, London SE23.

Monday, February 1

Chatting to Computers (6.30 p.m.) Dr C. R. Evans, Institution of Electrical Engineers, London Graduate and Student Section, at Savoy Place, London WC2.

Difficulties Inherent in the Study of Gas Exchange and Circulation of Blood in the Lungs (6 p.m.) Dr H. N. Duke, University of London, at the Royal Postgraduate Medical School, Du Cane Road, London W12.

Modern Electric Motor Protection (7 p.m.) Mr W. J. Johnson, Institution of Electrical Engineers, at the Royal Star Hotel, Maidstone, Kent.

Pre-Service Component Testing of Steam Power and Process (6.15 p.m.) Mr A. J. Morton, Institution of Mechanical Engineers, at the University of Durham.

Some Aspects of the Chemistry of Metal-Olefin Complexes (6.30 p.m.) Professor Sir Ronald Nyholm, FRS, Society of Chemical Industry, at the Scientific Societies Lecture Theatre, 23 Savile Row, London W1. (Jubilee Memorial Lecture.)

The Use of Instrumentation in the Evaluation of Paint Performance (7 p.m.) Mr D. M. Bishop, Oil and Colour Chemists' Association, at the Bullock Lecture Theatre, Hull College of Technology.

Reports and Publications

not included in the monthly Books Supplement

Great Britain and Ireland

PEP. Broadsheet No. 523: Changing Manpower Needs: a Study of Industrial Training Boards. By Santosh Mukherjee. Pp. iv+123. (London: Political and Economic Planning, 1970.) 24s. [712]

Ministry of Transport: Road Research Laboratory. RRL Report LR 368: The Use of Headlights in Lighted Streets. By L. Cooper. Pp. 34. (Crowthorne, Berkshire: Road Research Laboratory, 1970.) *Gratis*. [712]

Ministry of Agriculture, Fisheries and Food. Manual of Nutrition. Pp. v+106. (London: HMSO, 1970.) 9s 6d (47½p) net. [812]

New Possibilities and Techniques for Land Use and Related Surveys, with special reference to the Developing Countries. By D. P. Bickmore *et al.* (Papers presented at an International Symposium, London, 21–23 April 1970. The World Land Use Survey, Occasional Papers, No. 9.) Pp. viii+138. (Berkhamsted, Hertfordshire: Geographical Publications, Ltd., 1970.) 40s. [812]

Ministry of Transport: Road Research Laboratory. RRL Report LR 155: Comparative Head-On Impact Tests of Cars with either Front Transverse or Rear Mounted Engines. By J. G. Wall, R. N. Kemp and J. Harris. Pp. 119 (103 plates). (Crowthorne, Berkshire: Road Research Laboratory, 1970.) *Gratis*. [912]

Forestry Commission. Booklet No. 26: Metric Volume Ready Reckoner for Round Timber. Pp. 80. 7s (35p) net. Booklet No. 31: Metric Top Diameter Sawlog Tables. Pp. 23. 2s 6d (12½p) net. Leaflet No. 5: *Fomes annosus*: a Fungus Causing Butt Rot and Death of Conifers. Pp. 10. 3s (15p) net. Forest Record No. 76: Polecats. By Trevor B. Poole. Pp. 17. 2s 6d (12½p) net. Forest Record No. 77: Hedgehogs. By Patrick Morris. Pp. 18. 2s 6d (12½p) net. Census of Woodlands, 1965/1967: a Report on Britain's Forest Resources. Pp. 108. 12s (60p) net. Forest Record No. 31: Supplement No. 1 to Tariff Tables—Metric Tariff Tables. Pp. 10. 2s 6d (12½p) net. (London: HMSO, 1970.) [1112]

R & D Management, Vol. 1, No. 1, October 1970. Edited by A. W. Pearson. Pp. 1–48. (Oxford: Basil Blackwell, 1970.) [1112]

Other Countries

National Botanic Gardens of South Africa. Report 1969/1970. Pp. 56. (Kirstenbosch, Newlands, Cape Province: National Botanic Gardens of South Africa, 1970.) [312]

US Department of Health, Education and Welfare. Public Health Service—National Institutes of Health. Progress Against Cancer: a Report by the National Advisory Cancer Council. Pp. viii+98. (Washington, DC: Government Printing Office, 1970.) \$1.75. [312]

Centre National d'Études Spatiales. Rapport d'Activité, 1er Juillet 1969. Pp. 136. (Paris: Centre National d'Étude Spatiales, 1970.) [312]

Zoology Publications from Victoria University of Wellington. No. 52: Sea-Stars (Echinodermata: Asteroidea) from "El-tanin" Cruise 26, with a Review of the New Zealand Asterois Fauna. By Helen E. Shearburn Clark. Pp. 31. (Wellington: Victoria University of Wellington, 1970.) [412]

Australia: Commonwealth Scientific and Industrial Research Organization. Annual Report of the Division of Tropical Pastures, 1969/1970. Pp. 173. (Brisbane: CSIRO, 1970.) [712]

Science Council of Canada. Report No. 8: Seeing the Forest and the Trees—a Report on Forest Resources Research. Pp. 22. \$0.75. Report No. 9: This Land is Their Land—a Report on Fisheries and Wildlife Research in Canada. Pp. 41. \$0.75. (Ottawa: Queen's Printer, 1970.) [712]

Food and Agriculture Organization of the United Nations: The State of Food and Agriculture 1970: World Review; Review by Regions; Agriculture at the Threshold of the Second Development Decade. Pp. xii+274. (Rome: FAO; London: HMSO, 1970.) 60s; \$7.50. [712]

Norsk Polarinstitutt. Skriftr Nr. 153: Geologie und Fauna der Tertiären Ablagerungen Zentral-Spitsbergens. Von Klaus Vonderbank. Pp. 119+21 tafeln. (Oslo: Norsk Polarinstitutt, 1970.) [712]

Twenty-fifth Annual Report of the Council of the Queensland Institute of Medical Research for the year ended June 30, 1970. Pp. 23. (Brisbane: Queensland Institute of Medical Research, 1970.) [712]

Australia: Commonwealth Scientific and Industrial Research Organization. Annual Report of the Division of Irrigation Research 1969/1970. Pp. iv+77. (Griffith, NSW: CSIRO, 1970.) [712]

50 Jahre Forschungsförderung in Deutschland 1920/1970. Von Thomas Nipperdey und Ludwig Schmugge. Pp. 132. (Bonn-Bad Godesberg: Deutsche Forschungsgemeinschaft, 1970.) [712]

The Ecological Backlash—Nature Versus Man. By Professor J. M. Thomson. (Inaugural Lecture delivered at the University of Queensland, 18 June 1970.) Pp. 15. (St. Lucia, Queensland: University of Queensland Press; London International Scholarly Book Service, Inc., 1970.) 7s. [812]

Nederlandse Vereniging voor Weer-en Sterrenkunde. Observations of Variable Stars, January–June 1970. (Report No. 18.) Pp. 6. (Groningen, Netherlands: Kapteyn Astronomical Laboratory, 1970.) [812]

Harvard University Program on Technology and Society. Sixth Annual Report, 1969/1970. Pp. 103. (Cambridge, Mass.: Harvard University, 1970.) [812]

Bureau International des Poids et Mesures. Le Système International d'Unités (SI). Pp. 36. (Sèvres: Bureau International des Poids et Mesures, 1970.) [812]

Population Reference Bureau. Annual Report for 1969. Pp. 32. (Washington, DC: Population Reference Bureau, 1970.) [812]

Smithsonian Contributions to the Earth Sciences, No. 4: Volcanic Eruption at Metis Shoal, Tonga, 1967–1968—Description and Petrology. By William G. Melson, Eugene Jarosewich and Charles A. Lundquist. Pp. 18. \$0.30. Smithsonian Contributions to Zoology, No. 66: On the Ecology of the Caribbean Chitons *Acanthopleura granulata* Gmelin and *Chiton tuberculatus* Linné: Density, Mortality, Feeding, Reproduction and Growth. By Peter W. Glynn. Pp. 21. \$0.35. (Washington, DC: Smithsonian Institution Press, 1970. For sale by US Government Printing Office.) [812]

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